MASTER IN BIOMEDICINE

Assessing the impact of control action plans in the prevention and control of cystic echinococcosis in Morocco

KIDANEMARIAM, Meaza

Award date:
2023

Awarding institution:
University of Namur

Faculté de Médecine

Assessing the impact of control action plans in the prevention and control of cystic echinococcosis in Morocco

Mémoire présenté pour l'obtention
du grade académique de master en sciences biomédicales (Master 60)

KIDANEMARIAM Meaza

August 2023

Université de Namur
FACULTE DE MEDECINE
Secrétariat du département de Sciences Biomédicales
Rue de Bruxelles 61 - 5000 NAMUR
Téléphone : + 32(0)81.72.43.22
E-Mail: manon.chatillon@unamur.be - http://www.unamur.be/

Assessing the impact of control action plans in the prevention and control of cystic echinococcosis in Morocco

KIDANEMARIAM Meaza

Abstract

Cystic echinococcosis (CE) also known as hydatid disease is a zoonotic parasitic infection that is reported by World health organization (WHO) as one of 20 neglected diseases worldwide. The disease is endemic in many regions of Morocco. It is more prominent in regions where animal husbandry is commonly practiced. The objective of this study is to understand which control measures have been implemented since the initiation of the national hydatid control programs and to identify areas of improvements. The research method used for this study is a thorough literature review from related publications found in electronic databases such as PubMed, google scholar and guidelines. Data unrelated to this study's aim was used as exclusion criteria. Articles that are written in English and linked to the study were used as inclusion criteria. Effective control measures against cystic echinococcosis depends on several factors including epidemiological state of the country, geographical distribution, strength of policies and infrastructure set up of the country. It is important to put into consideration the preparation of management guidelines that contain tailored control measures in accordance with the specific country planned to be implemented.

Key words: Cystic echinococcosis, control measures, Morocco

Mémoire de master en sciences biomédicales (Master 60)
August 2023
Thesis Supervisor: Professor Nathalie KIRSCHVINK (URPhyM)

Assessing the impact of control action plans in the prevention and control of cystic echinococcosis in Morocco

KIDANEMARIAM Meaza

Master's student in Biomedical Sciences- Université de Namur

Word count: 11,100
Figure count:5

Acknowledgment

I wish to take this opportunity to express my heartfelt thanks to all the individuals who facilitated in the fulfilment of this work.

To my thesis supervisor: Professor Nathalie KIRSCHVINK, thank you for agreeing to supervise me during this research project. I am grateful for the encouragement, support, and time you dedicated to me throughout this work.

I also like to thank Professor Jean-Pierre GILLET for the providing me guidance during the master's study in times of need.

I would also like to thank members of the jury for taking time to evaluate this work.

I am also grateful to my parents and siblings who have been my support system throughout my education and career life.

Table of content

List of Figures

1. Introduction

1.1. Background

Cystic echinococcosis, also known as hydatidosis is been a major public health burden. It is a zoonotic disease that has a worldwide distribution of 1-200 cases per 100000 annual incidence rates. It is a parasitic infection with the etiologic agent of larval cystic stage of the taeniid tapeworm. It causes pathology to humans through direct close contact with dogs and herbivorous animals by consumption of parasitic eggs during grazing (1).

Morocco having a vast demography and larger surface area in northern Africa, contributes to the major share of the overall *cystic echinococcosis* burden in the North Africa/middle east region. The overall dog population in Morocco accounts for around 2 million. Most are free-roaming owned dogs and stray dogs (2), with an infection rate in Middle Atlas Morocco ranging from 23.5% to 38.8% and from 51.3% to 68.5% respectively (3).

There is a scattered distribution of cystic echinococcus infection rate in Morocco. Certain regions are reported to have a higher rate of infection than some of the other regions. Factors such as lack of infrastructure mandatory for proper surveillance become challenging to implement. This results in underestimation of cases and delayed public health response (4).

It is believed that there is underreporting of the disease. This is because humans barely experience symptoms of illness and confirmation of disease relies mostly on laboratory testing and diagnostic modalities such as ultrasound and CT scans. The availability of these investigating modalities is limited in resource-limited regions of the country.

There is also the issue of the presence of numerous amounts of unregulated slaughterhouses in the country. In addition, home or village slaughtering of sheep for cultural and religious festivities are very common which results in unreported cases of *Echinococcus* infection (2).

A varying degree of knowledge among different sectors such as butchers and health professionals has increased the likelihood of transmission. Limited understanding of the mode of transmission and lifecycle of disease prevents the inhabitants from applying necessary preventive practices (5).

1.2. Significance of the study

Close interaction between humans, animals, and wildlife has been a detrimental factor in the transmission of most zoonotic infections including *Echinococcosis granulosus*. Moreover, socioeconomic factors including limited resources and health care, and inadequate sanitation have contributed to challenges faced in endemic areas. To assess the effectiveness of different control measures in reducing the incidence and prevalence of *Echinococcus* numerous studies have been conducted throughout the years. Despite such efforts, there are significant discrepancies and gaps in the outcome of implemented programs across different countries. The study aims to address these gaps with a particular focus on Morocco and to compare with the measures applied worldwide.

Morocco is one of the countries impacted by *Echinococcus granulosis* infection. The infection rate remains high despite the implementation of various control measures. This is particularly common in the rural areas where livestock farming is practiced. The study will emphasize on Morocco to find the reason as to why certain measures that were taken have failed and which control action plans or combinations of them had shown promising results in reducing the incidence of the disease in various regions of the country. In addition, the purpose of this study will be to provide evidence-based recommendations that were tailored as effective strategies in the management of *Echinococcus granulosus* infection worldwide.

1.3 Literature Review

1.3.1. General overview

Echinococcosis is one of the neglected zoonotic infections. The disease is considered neglected for reasons that it mostly occurs in resource limited settings leading to under-reporting and limited awareness by responsible sectors, thereby masking the true burden of the disease. Echinococcosis is one of the 20 disorders on the WHO's list of neglected diseases that need better global health care provision. Humans, animals, and the environment contribute to the transmission and maintenance of the disease. Its interdisciplinary nature and involvement of multiple hosts make it challenging to control and requires an integrated approach (6).

1.3.2. Types of *Echinococcus* species

Echinococcus belongs to the genus family of Taenia. The adult parasite is around 7 mm long. Its morphology is made of head also called scolex that has hooks and suckers, the body strobilla composed of many segments known as proglottids. The life cycle of *Echinococcus* includes a definitive host that is mostly dogs and wild carnivorous animals and an intermediate host that are livestock. There are substantial differences between the types of *Echinococcus* species. Such heterogenicity is seen in terms of morphology, host preference, lifecycle and transmission pattern, disease progression rate, and geographical distribution. It is also seen in the location of cysts and organs it involves, diagnostic specificity and sensitivity, and response to treatment options. It is highly relevant to know such variability because it provides a stepping stone in the management and control of the disease.

Echinococcosis is caused by various species. *E. granulosus* and *E. multilocularis* are the causative agent for *Cystic echinococcosis (CE)* or *hydatidosis* and *Alveolar echinococcosis (AE)* respectively that are common zoonotic infections of humans. *E. equinus, E. ortleppi,* and *E. Shiquicus* have a minor capacity to induce a public health threat compared to the well-known pathogenic species, *E. granulosus* and *E. multilocularis*. *E.oligarthrus* and *E. Vogeli* are less frequently seen but when they occur, they cause *Polycystic Echinococcosis (PE)* and *Uniocular Echinococosus (UE),* respectively (CDC, 2019). The disease is due to the larval stages of cestodes which is a member of the genus *Echinococcus* (7).

E. multilocularis causes *Alveolar Echinococcosis (AE)* mostly infecting animals particularly smaller mammals such as foxes. Some mice species are the intermediate host and humans become accidental intermediate host. It is mostly observed in countries located in the Northern Hemisphere such as Canada, the USA, Europe (Germany, Austria, France, and Switzerland), Japan, and Russia. Cross-country trading of animals and animal products and certain human activities such as hunting has made control measures to be ineffective in halting the disease incidence (6).

The prevalence of alveolar *echinococcosis (AE)* in Europe and the northern hemisphere is associated with the influence of globalization. The introduction of wild animals living close to urban villages accompanied by human living expansion and formation of new settlements contributed to the disease's existence and spread. Compared to other wild animal species, red foxes show a higher zoonotic potential and host specificity to *echinococcosus multilocularis* which causes *alveolar echinococcosis (AE*) (8).

E. Oligarthrus has a similar disease presentation as cystic echinococcosis but is rarely seen in humans and mostly observed in wild animals and rodents. *E.vogeli* has a low incidence but when it does occur it leads to the formation of *polycystic echinococcosis* (9).

1.3.3. Lifecycle of *Echinococcosus granulosus*

The life cycle of *Echinococcus granulosus* starts in the intestine of dogs or other canids which are the definitive hosts. The definitive host consumes the organs of the intermediate host infected with the parasite larvae. The adult worms produce eggs inside the intestine which will be released through faeces. Once the egg is released into the environment it can endure numerous conditions such as dryness, low temperatures, soil type (highly acidic or alkaline), and UV exposure until it finds a suitable host due to its thick keratinized outermost layer. It can last up to several months up to one year.

As shown in the figure below the adult worm (**1**) is 3 to 8 mm in length and it consists of a scolex (head) and three proglottids (segments). The neck connects the scolex and the proglottids. The scolex has suckers and hooks that serve to attach to the wall of the intestine (**6)** of the definitive host. The proglottids are anterior, middle, and posterior (terminal). The anterior proglottid is immature as it has underdeveloped reproductive organs. The middle proglottid is mature and has fully developed reproductive organs, and the posterior proglottid is also called gravid since it has fully developed eggs ready to be expelled into the environment. There are different developmental stages of the parasite in the definitive host which takes around 45 days. These stages are essential to understand its pathogenesis, and selection of different diagnostic tests. In addition, in terms of treatment, these various stages respond to treatment differently. Day 1 corresponds to evagination and elongation of the protoscolex.

From day 11 to 14 the formation of the first proglottid occurs. Following that between day 14 and 17, both male and female reproductive structures start to form. Then, between days 17 to 20, the addition of new proglottids to its body occurs through segmentation. On the days between 20 to 28 days male structures have fully formed while the female structures remain developing. Between days 28-33 both male and female reproductive organs reach full development. From days 33-37, the proglottid at the end gets fertilized, and between days 37-45, the last segment is filled with eggs and ready to be released into the environment seen in fig 2 below (2).

The intermediate hosts livestock (cattle, sheep, pigs etc...) consume the egg while grazing. Once ingested, the digestive enzymes in the gut facilitate the eggs to hatch and to release oncospheres (10). Certain secretions of the oncosphere and its six hook-like structures allow it to penetrate the intestinal mucosa **(3)**. This is an essential step in the initiation of the infectious process. Once in the bloodstream, it starts to infect various organs such as the liver, and lungs forming hydatic cysts also known as the metacestode stage seen in **(4)**. It takes one to two years for the hydatid cysts to develop and become detectable in the livestock. The hydatid cysts comprise of a thick outer layer and an inner layer with germinal cells. The germinal cells, capable of sexual and asexual reproduction give rise to brood capsules that produce thousands of protoscoleces **(5)** (11).

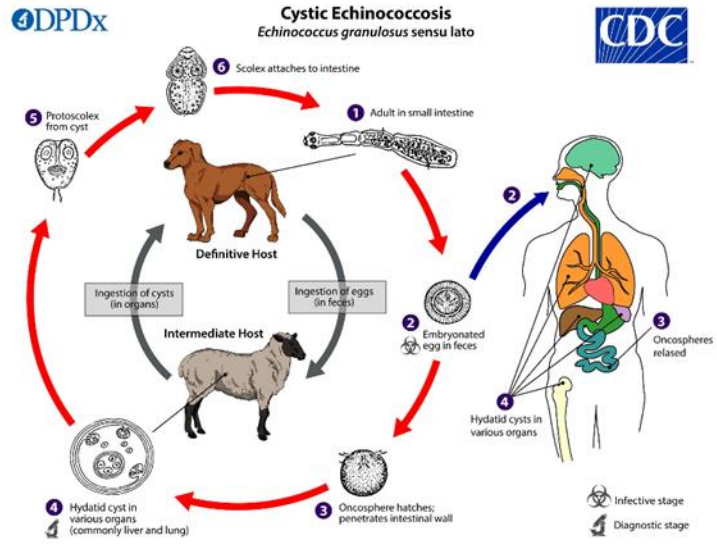

Fig. 1 Life cycle of *Echinocosus granulosus*
https://www.cdc.gov/parasites/echinococcosis/biology.html

Here in the intermediate host, some of the metacestodes will produce protoscoleces through asexual reproduction. Dogs ingest organs (liver, lungs) infected with cysts. Once the definitive host ingests the cysts, protoscoleces found within the cysts undergo sexual reproduction. By undergoing different developmental stages in the small intestine of dogs (fig 2), the prostoscolex develops into mature adult worms. It takes about 32 to 80 days to form into the adult stage. The adult worm attaches to the small intestine using the scolex. *E.multiocularis*, another common species of *echinococcus*, exhibits similar developmental stages as seen in figure 2. The structure of the scolex includes four suckers and a crown of hooks which makes it resistant to the intestine peristaltic movement allowing it to release eggs.

The protoscoleces can sustain the external environment and play a crucial role in the parasite pathogenicity. Humans get infected through direct contact with the definitive host in most cases from domestic dogs (12).

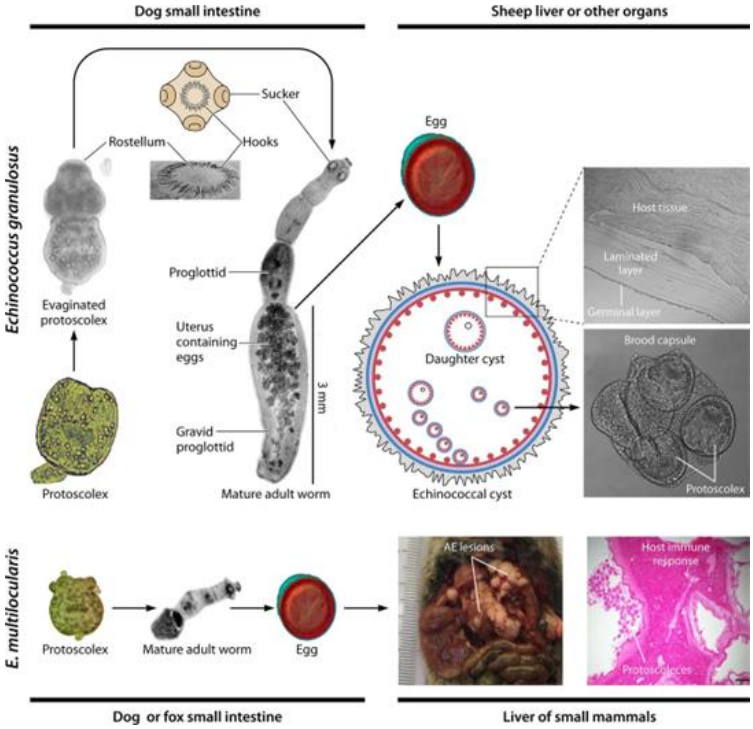

Figure 2. Overview of the different developmental stages of *E.granulosus* & *E. multiocularis* https://journals.asm.org/doi/10.1128/cmr.00075-18

The definitive hosts for *Echinococcus granulosus* are primarily domestic dogs. However, in regions where wild carnivores are in abundance or habitual factors such as hunting is common, humans become infected by foxes. In humans, the ingested eggs hatch in the bowel and enter the blood circulation. The metacestode stage, the infective stage of *Echinococcus granulosus* in humans causes varying degrees of symptoms. In most cases, patients are asymptomatic. The symptoms depend on the organ's involvement. The liver, mostly the right lobe (70%) and lung (20%) are common sites. In 40-80% of the cases, patients will have one organ infected that contains a solitary cyst (13).

The cyst contains transparent fluid. The growth of cysts can vary (estimated time would be 1–30 mm per year). Cysts can rupture and release the contents of fluid, membranes, and daughter cysts (fig 2). This causes metastatic infection and organ failure. Therefore, it requires immediate treatment to prevent fatal complications. Likewise, the daughter cysts contain larvae and are

responsible for the primary cysts' growth. This occurs when the daughter cysts grow by consuming nutrients from the host. It subsequently increases the size of primary cyst which can have a pressure effect on organs of the host. On the other hand, the calcification of an echinococcal cyst occurs, and the fluid inside the cyst hardens with time. These are detected by diagnostic imaging techniques. Several factors impact the clinical presentation of the disease. These include the nature, size, and location of the cysts, the degree of organ involvement, and the immune status of the individual. Since the incubation period takes an exceedingly long time in humans, patients with *cystic echinococcosis* can be symptom-free for months or years. Calcified cysts rarely cause infectious-like symptoms, however if large enough they can impose pressure on organs (14). *Echinococcus granulosus* is well known for its genetic heterogeneity having multiple strains within a population. The genotypes are encased in the *E. granulosus sensu lato* genotype complex that includes further genetic variants *E. granulosus sensu stricto* represents G1, G2, G3 genotypes, *E. ortleppi* G5 and the *E. Canadensis* comprises of G6, G7, G8, and G10 (15).

1.3.4. Genetic variability of *Echinocosus granulosus* species and pathogenic implications

Geographical distribution also has a certain impact on heterogenicity. The genotypes of *Echinococcus granulosus* can selectively be found in different regions more commonly than others. This geographical isolation can cause genetic drifting through time. For instance, a study showed *E.granulosus s.s* was found commonly in southern Europe, *E.ortleppi* in central and western Europe and *E. canadensis (G6/7)* was found in Eastern and central Europe (16). On the other hand, the immune response of the definitive and intermediate host and the life cycle's complexity. It is essential to understand such factors as they are the key in the management and control of *Echinococcus granulosus* infection (17).

Echinococcosus granulosus show strain diversity with specific genotypes isolated from various animals; G1 and G2 strains identified in sheep, G3 in buffalo, G4 in a horse, G5 for cattle (58), G6 camel strain, pig strain (G7) (18), cervid strain (G8) (19), etc. Such strain identification has epidemiological relevance. It is also linked to the degree of infectivity in humans, the preference of hosts in the lifecycle pattern, the determination of diagnostic modalities, and in the formation of effective vaccines (7).

In Mexico, two genotypes were isolated and found to be prevalent: in sheep G1 and in pig G7. G1 was causing widespread infection in humans and causing significant public health implications

(20). The transmission of *cystic echinococcosis* in Mexico is due to dog to domestic pigs transmission. This is mostly observed in rural areas where livestock farming (pigs) is common (21). A study was conducted in various regions of Peru to determine the genetic variations of *Echinoccosus granulosus*. The DNA sequence of genes was performed from 71 samples gathered from livestock. The result showed the G7 genotype (*E. canadensis* pig strain). Moreover, G1 was responsible for most of the infections in humans, sheep, and cattle. Sporadic findings of G6 (*E. canadensis* camel strain) were obtained in goats and humans (22).

In North Africa, there is intraspecies variability across different countries. Tunisia is considered as one of the countries that are hyper endemic to *cystic echinococcosis*. Most dogs have not received deworming medications.Various degrees of infection have been observed in the livestock population of sheep, cattle, dromedaries, and goats. The commonly identified genotype strains are G1, G3, G4, and G6, where G1 and G6 are notably prevalent (23).

According to various studies, Middle East and Western Asia have higher levels of intraspecific diversity than the rest of the world. What is peculiar about the diversity of *E. granulosus* s.s. isolates in these regions is that the genetic diversity is not only high among different populations of the same genotype but also within individual cysts, indicating the presence of multiple strains or subtypes of the parasite in the region (24).

A study conducted in Algeria found that livestock population was affected by two genotypes of the sheep strain and was identified in cattle and ovine. This was also evidently seen in humans infected with *cystic echinococcosis* who sought treatment. G6 was also the other common strain that was obtained in samples collected from African camels in this region (25).

According to a systemic review done within the time frame of (2000 –2021), Italy and Spain account for the largest numbers of *cystic echinococcosis* in humans in Europe (16). In Spain, various strains of *Echinococcus granulosus* with different pathogenicity were reported. This has practical implications in terms of the formation and implementation of treatment strategies. Two specific strains from Spanish pigs: G1 (sheep-dog strain) and G7 (pig-dog strain) genotypes were linked with significant health consequences in humans (26).

France, Poland, and Switzerland reported seven human cases of *cystic echinococcosis*. A solitary case of *cystic echinococcosis* was reported in Germany. Similarly, isolated case of the G10 strain of *Echinococosus granulosus* was identified in Finland and 4 cases of *cystic echinococcosis* in humans were found in China, Far East Russia, and Mongolia. In Europe, little or no contact

between humans and wild animal populations has been the main factor as to why human infection rates with these strains (G8 and G 10) are low.

In addition, in Europe, isolated human settlements and living in less proximity to the wild animal populations has been the contributing factor as to why human infection rates with these strains (G8 and G10) are low. This makes the parasite remain within the natural cycle in the wildlife. This is not the case with G1 and G3 strains, that are found in the livestock population and higher incidence rates in humans (16). G1-G3 and G6 genotypes are prevalent in Libya. An increase in the risk of transmission is evident in different regions of Libya due to close relationships with dogs. Household dogs that are not properly dewormed are engaged in farming and herding activities which pose a risk. Similarly, stray dogs live close to humans scavenging remnants of animal products (27).

1.3.5. Clinical symptoms, diagnosis, and detection of *Echinococcus granulosus*

In Northeast Africa, in Egypt G6 strain, and Sudan G5, G6-G7 strain are linked with *cystic echinococcosis* in humans as evidenced by molecular analysis of infected samples seen in figure 2 (Geographical distribution of the different genotype strains of *Echinococcus granulosus* across the world). Patients present symptoms like abdominal pain, distention vomiting, and in some cases cholestatic jaundice when the infection involves the liver. In 5-15% of the cases, they present pulmonary symptoms such as cough, chest pain, hemoptysis, and shortness of breath. In rare cases the brain, bone, ovary eye, etc. could be involved (28). The degree of contamination of the environment with parasitic eggs and the age of the host is highly associated with *Echinococcus granulosus* infection of the intermediate hosts (29).

Human cases of the disease can be detected using imaging modalities such as ultrasound, computed tomography (CT scanning), and magnetic resonance imaging. The disease is found in rural parts of endemic regions mostly with low infrastructure. Therefore, the availability of such imaging techniques is significantly challenging due to the limitation in their accessibility. Ultrasound has certain advantages in terms of availability, and accessibility and has public health importance by providing guidance for the diagnosis and treatment through WHO classification of *cystic echinococcosis*. Although a CT scan shows thorough images of cystic lesions and is mostly preferred for planned surgical treatment (10).

In the context of epidemiology and disease surveillance antibody detection against *E.granulosus* antigens through immunodiagnostics such as Enzyme-Linked Immunosorbent Assay (ELISA) have shown many advantages. It allows early detection of infection even before cysts appear by detecting circulating antibodies. It also assists in the monitoring, evaluation, and post-therapeutic follow-up of the efficiency of treatments. Moreover, it is suitable to be used in resource-limited facility setups due to its low-cost techniques (7).

The protoscolex , the cyst germinal cells and membranes, adult worms and oncospheres can be used to determine the DNA sequence of *Echinococcus granulosus* using PCR (30).

Evaluating the DNA sequence data using phylogenetic analyses determines various strains and subtypes of *Echinococcosus granulosus* and showcases the evolutionary relationships within the different species (31). Many factors are associated with the genetic variance of *Echinococcosus granulosus*. One of the reasons being during reproduction, *Echinococcosus granulosus* can have genetic recombination. This commonly occurs when various strains infect the same host and then their genetic material combines at the time of reproduction. This causes the development of new genotypes with higher levels of infectivity. A study performed in various countries of America evaluated the genetic variability and phylogeography of Echinococcosis G1(32). Such variability was determined by sequencing the mitochondrial genome. The result revealed high haplotype diversity.

WHO-IWGE PNM system classifies *cystic echinococcosis (CE)* into CE1-CE5 using imaging techniques. Within this classification CE1-5 considered active cysts with a simple fluid-filled cavity and no daughter cysts (CE1), cysts with daughter cysts and shows degeneration are (CE2) and cysts that are inactive with some degree of collapsed wall and a content that is solid in nature is recognized as (CE3). Inactive cysts with a calcified wall are taken as CE4 and with a partially or completely collapsed cavity and thickened calcified wall being CE 5. Furthermore, in 2020 a new category that includes CE6 or cystic lesion (CL) that have complex cysts with multiple compartments, septations, and components that are solidified which do not fit the initial classification (33).

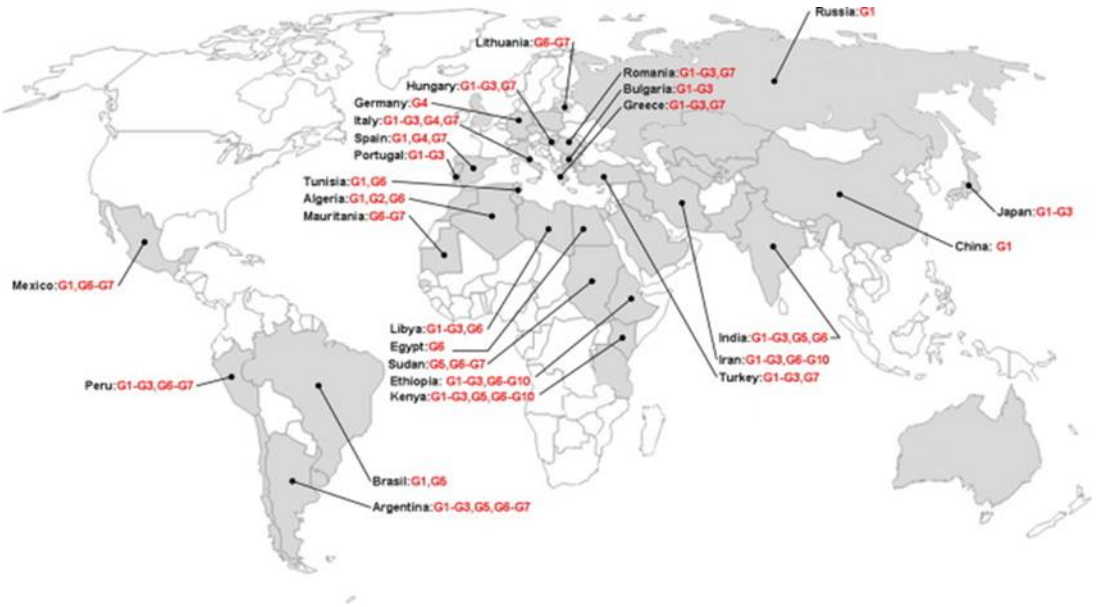

Fig 3; geographical distribution of the different genotype strain of *Echinococcosus granulosus* https://doi.org/10.1016/j.vetpar.2012.09.027

1.3.6. Worldwide distribution of *cystic echinococcosis*

E. granulosus is globally widespread with a diverse variation in epidemiological distribution. In western Mediterranean countries such as Spain and in regions located in the Northeastern, Central, and Western parts of the country, high incidence rates ranging from 1.1 to 3.4 cases per 100,000 inhabitants has been reported (34).

WHO stated that among humans 50 per 100,000 persons every year get affected by cystic echinococcosis in endemic areas. The prevalence can reach as much as 5% to 10% in regions such as Argentina, Peru, East Africa, Central Asia, and China. Slaughterhouses in South American regions where the incidence rate is intensive have a varying degree of prevalence of 20-95% (6).

According to the Europe surveillance report in 2020 on *Echinococcus granulosus* 529 cases of echinococcosis were found. Out of these 243 cases are caused by *Echinococcus granulosus*. the number of confirmed cases of *echinococcosis* in 2020 in different countries of EU are Austria (34 cases), Belgium (19 cases), Bulgaria (95 cases), France (53 cases) and Germany (152 cases). In Finland and Italy no confirmed cases were reported during this year (35).

A meta-analysis done on 13 countries in 5 sub-African regions showed an overall prevalence of 1.7% in human *Echinococcus* infection. The highest prevalence was observed in Eastern Africa (2.7%) with Sudan having the highest prevalence rate (49.6%) (36). *Cystic echinococcosis* is

endemic in Morocco with overall prevalence of 1.9%. It has variability among the different regions. Rural areas such as Ifrane have higher rates (2.6%) than El Hajeb (1.3%) (4). In other regions such as Loukkos and Tiznit a considerable proportion of dogs were infected with *E. granulosus* with mean prevalence rates of 58.82% and 55.42% respectively showing higher disease burden in these regions (37).

1.3.7. Economic impact of *cystic echinococcosis*

Economic and financial loss associated with *cystic echinococcosis* is significant with 1-3 million disability-adjusted life years per year. The cost of treating patients is dependent upon the average estimated cost of living of the region and individual/family income (6). Equipped hospital staff and the wages of medical professionals can drive up the expenses associated with *cystic echinococcosis* treatment. In addition, variations in the health care systems and regulations across countries also determine the value of importance given to surveillance and control of the disease. Such health care disparities impose negative public health implications because it becomes impossible to include innovative approaches to provide sustainable interventions such as improved diagnostic tests, facilitate research and engage health care staff in training. The paper described the disease's socioeconomic impact in terms of disability-adjusted life years (DALYs) and monetary losses incurred in both human and livestock populations, based on previous case reports (38).

According to Budke, an estimation of 285,407 disability-adjusted life years (DALYs) or an annual monetary loss of US $193,529,740 was reported. Similarly, an annual loss of US 141,605,195 US in livestock production was reported. It also mentioned that there is a considerable amount (nearly 4-fold degree) of underreported cases due to several factors. These are due to patients infected with *cystic echinococcosis* in the first few years who show no symptoms or vague symptoms that are nonspecific to *cystic echinococcosis*. Rural areas located a great distance from healthcare facilities are left out of surveillance and data collection. Furthermore, cultural, and religious factors also can impact individuals to seek traditional treatment methods instead of hospital visits (38).

1.3.8. Treatment options of *Echinococcosus granulosus* infection

Surgery and chemotherapy are the two forms of treatment modalities for *cystic echinococcosis*. Patients present with mass effects from large cysts can benefit from surgery. Moreover, infected cysts and cysts that are easily accessible are amenable to such treatment.

Albendazole and mebendazole are benzimidazole compounds that are widely used to treat *cystic echinococcosis*. The efficacy of these medications can be assessed based on the decrement in the viability of cysts, which improves as the treatment is consistently taken for a longer duration. With one month of treatment, 72% of cysts become non-viable whereas 94% with 3 months of treatment. Studies have shown that co-administration of praziquantel with albendazole has been more effective. This is linked to the ability of praziquantel to increase the mean maximum plasma concentration of albendazole by increasing albendazole sulfoxide levels in the bloodstream (7).

The intermediate hosts can act as reservoir hosts. EG95 vaccine is a recombinant vaccine that is found to provide a (96–100%) protection against *echinococcosus granulosus*. Eg95 vaccine 50 μg with a particular adjuvant Quil A (Brentag Nordic) of 1mg for sheep and goats has been formulated in Australia by a team of researchers led by Dr. Marshall Lightowlers at the University of Melbourne (39). The vaccine is administered in two initial doses given several weeks apart, followed by annual booster shots to maintain immunity.

Following vaccination antibody mediated and complement mediated lysis of the oncospheres occurs which represents the parasitic stage that vigorously spreads in the intermediate hosts.

It also decreases the duration required to implement control strategies efficiently. One of the advantages of the EG95 vaccine is that it can be produced at a broader scale and cost-effective way. Anthelmintic treatment of dogs alone is not sufficient to eradicate E. granulosus in most countries. Therefore, vaccination of these hosts along with dog chemotherapy minimize infection in dogs subsequently lower infection risk for humans (7). Several countries, including Australia, New Zealand, Argentina, Chile, and Peru have given the licensed vaccine to be used as part of the control measures of *Echinococcosus granulosus* infection transmission.

According to serologic and necropsy results in Rio Negro, Argentina marked a reduction in the prevalence rates between unvaccinated sheep (66%) and vaccinated sheep (21%). Similarly in postmortem examination, cysts were found in only 7% of the vaccinated goats whereas same examination revealed 66% among unvaccinated goats (40).

According to the WHO ultrasound images classification of *Echinoccosus granulosus* cysts, treatment strategies in humans are recommended. As seen in fig 4, surgical removal of cyst in combination with albendazole treatment is treatment of choice for CE 2 and CE 3b that are >5cm. Isolated treatment with albendazole is recommended for CE1 and CE2 <5cm, whereas coupling albendazole with PAIR (puncture aspiration injection reaspiration) which is a non-invasive alternative to surgery is suggested for CE 1 and CE3b > 5cm. Evacuation of the content of the cysts using MoCAT (Modified catheterization technique) coupled with albendazole is recommended for CE 2 and CE 3 that are < 10 cm. For biologically inactive cysts that are it is possible to wait and see with a given follow up.

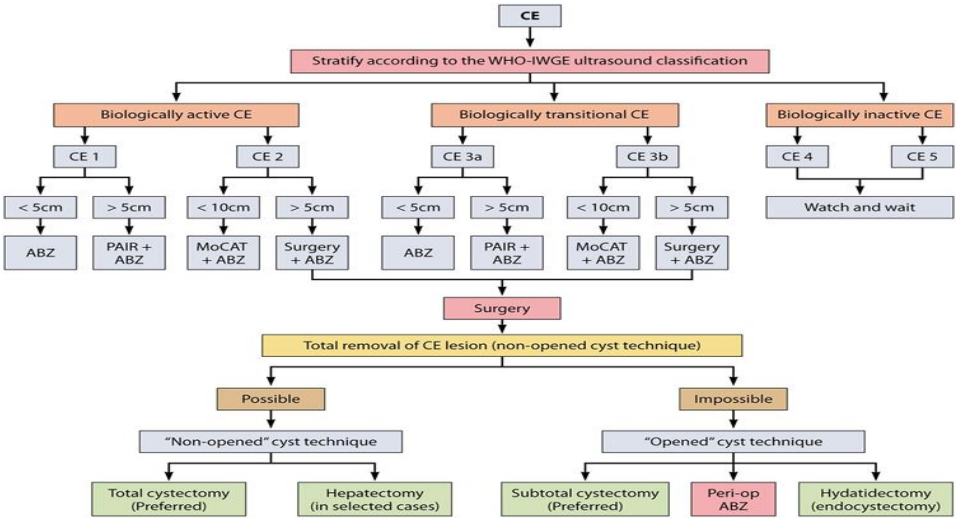

Fig. 4 Treatment protocol for *cystic echinococcosis* (CE) on the basis of WHO-IWGE international classification of ultrasound images of cysts. https://journals.asm.org/doi/10.1128/cmr.00075-18

1.3.9. Comparative assessment of control measures across different countries

Iceland was the first country to implement hydatic control programs. Such control measures included deworming of dogs surveillance of slaughterhouses, and mass screening of sheep population. This has resulted in the complete eradication of the disease. For 15-50 years such measures have been improved and applied by other Iceland countries Tasmania, Falklands, and

Cyprus, and were found successful. The administration of praziquantel in combination with the usage of insular models was found effective in reducing human incidence rates (41).

In South America, new control programs were launched in endemic areas that involved the administration of praziquantel 5mg/kg eight times a year. The aim was to cease the parasite to get into the gravid stage and prevent the release of eggs. The lack of infrastructure and the movement of dogs during winter was one of the challenges that hindered such measures. Uruguay has a successful track among South American countries. Initially Abdominal ultrasound screening examination revealed a total prevalence of human cystic echinococcosis cases at 5.6% 64 out of 1,149 (57). Since the start of deworming of dogs in Uruguay in 1995, the prevalence of dogs has tremendously declined to 0.7%. and human incidence rate to 6.5/100 000 (42). Praziquantel treatment of domestic dogs, elimination of stray dogs to some extent in conjunction with health education and surveillance with assigned village hydatid disease officer in Hutubi and Wensu Xinjiang China was implemented for four consecutive years. This resulted in the reduction of the prevalence rate in these regions to 14.7% and 18.6% respectively, and subsequently to zero.

Obtaining the infection densities in the definitive and intermediate hosts allows a targeted approach to the management of *cystic echinococcosis*. There are several endogenous and exogenous socio-ecological factors contributing to increased infection intensity and prevalence in the hosts. Environmental factors such as climatic variations, land usage, source of water, and the percentage of areas covered by vegetation are considered endogenous factors. Climatic variations determine parasite survival, and vegetation coverage influences the dispersion of the intermediate hosts cattle and sheep. Exogenous factors including age, sex, and feeding behaviors of the hosts are also contributing factors to the intensity of infection. For example, female dogs and pups tend to have the stress of reproduction and underdeveloped immune systems respectively leading to an increased rate of infectivity (43).

In the framework of hydatid control programs 5 Iceland countries such as New Zealand, Cyprus, Tasmania, etc. a two-way approach “horizontal” and “vertical “was implemented. The horizontal approach gave emphasis on education about the disease and improving health awareness together with meat inspection and improving slaughterhouses. The vertical approach included setting up guidelines on the strategies that apply to veterinary public health measures, primarily targeting dogs, also called a fast-track approach. Reducing the number of non-owned dogs, frequent testing

and treatment of owned dogs was applied. A descriptive approach of the two approaches led to complete and nearly complete eradication of human hydatidosis in these countries.

These two combined approaches that were applied previously in New Zealand were somewhat implemented in Sichuan province of China with additional measure taken to preventing livestock to go into mountains for consecutive six months. The six months window period is attributed to the seasons of harsh winter. During this period, vegetation covers most of the land and many livestock populations are out for grazing. People in this region follow buddha religion and believe that when an animal die, it should be left undisturbed. This practice allows numerous stray dogs to scavenge the carcasses. Through time this was recognized by the community and prevented their herds from being exposed. This action worked well in favor of the proposed hydatic control program aimed at dogs. However, in far rural regions supervised administration of antihelmenthics was difficult (44).

Measures taken in Tibetan communities, in China, were somewhat different. At the outset, it focused solely on dog management. Emphasis was given to "roaming behavior," regular deworming "and "change feeding habits." These measures applied were found to be effective in reducing the disease's prevalence to a lower rate. The expected objective was to reduce the prevalence of *cystic echinococcosis* in dogs to 5% in 2020. These consist of reducing the number of stray dogs, frequent deworming within a year (8 times a year) and alternating the feed of dogs from discarded viscera to regular human food. Households strictly implemented leashing their dogs throughout 24 hours. This method and strategy also highlighted the prevention of re-infection of dogs and breaking the life cycle of *E. granulosus* by restricting contact of dogs with livestock which ultimately reduces the risk of transmission to humans. It is important to take into consideration different social, cultural and religious factors in implementing different types of control measures of *cystic echinococcosis*. For example, a control measure that was implemented in China was getting rid of excess stray dogs. This is not applicable in regions with Buddhism is practiced as a religion as dogs have spiritual value (45).

In the context of Morocco there are several factors that should be put into consideration in order to implement most of the observed strategies in different countries.

2. Aim of the Study

2.1. General objective

- Recognize and have a better understanding of the significant burden posed by *E. granulosus* infection and to improve public health outcomes in Morocco.

2.2. Specific objectives

- To identify areas for improvement- reveal areas where the control measures could be improved or optimized by identifying gaps in the implementation or enforcement of the control measures, which could be addressed through improved coordination and communication between relevant stakeholders.
- To inform future planning- To make evidence-based decision-making and provide valuable information for the development of effective control measures of *Echinococcosus granulosus* infection and provide guidance on the formation of future initiatives.

3. Barriers in attaining effective control measures in Morocco and in different countries

3.1 Possibility of re infection

In addition, specific factors have made the control measures of *cystic echinococcosis* in Morocco to be unsatisfactory. The reinfection of dogs is one of the common reasons. The slaughtering of livestock is mostly done inside a community or in each household at times. Lack of proper disposal of offal allows dogs access to dead animals organs. Dogs that consume will be predisposed to reinfection. Similarly, government or private organizations that run the slaughterhouses lack sufficient surveillance and do not apply adequate biosecurity measures which facilitates the risk of transmission.

A study was done in Morocco to show regional variations in the prevalence of *cystic echinococcosis* in different farm animals. Such variations were influenced by the density of population of dogs, the degree of dog's engagement in people's lives and commitment to public health surveillance. The five regions (Rif, Loukkos, mountains of middle Atlas, region of Rabat and Casablanca and the southern part of Morocco) selected for the study seen in the fig 3. exhibit different geographical and climate set up, mode of living and human activities. The middle atlas

is the mountainous region of Morocco. The localities depend mostly on livestock farming. This place was found to have the highest prevalence of CE in cattle (48.72%), in sheep (11.14%) and in goats (11.14%). This was attributed to the considerable number of dog population which serve as the reservoir of adult parasite and egg shedding.

The northern part of Morocco Rif, extending to the Mediterranean coast showed a prevalence of (6.20%) in cattle while (5.12%) and (0.03%) in sheep and goats was observed respectively. The topographical arrangement and livestock farming practices in this region established a less favorable environment for the transmission dynamics than Middle Atlas. Southern part of Morocco has a prevalence of (13.33%) in cattle, (1.42%) in sheep, in goats (2.19%) and camels (10.87%). In Loukkois significantly high prevalence rate of *cystic echinococcosis* was seen in cattle (37.61%) accompanied by 31.65% in sheep and 0.00% in goats (2).

Fig 5 Farm animals hydatidosis study areas
https://www.sciencedirect.com/science/article/pii/S0304401706000185#aep-section-id10

Another study was undertaken to assess dog chemotherapy in Morocco. Three villages labelled (Group A, Group B, and Group C) were selected in Ifrane province that are located only 20-30 km far apart from each other. Their resemblance in most aspects such as human activities, herd population and climatic conditions was considered in selection of the villages. The impact of different time intervals of chemotherapy was used as the only determining factor to evaluate the risk of infestation in dogs. The three villages with dogs of Group A, Group B, and Group C received praziquantel three times at intervals of two months, three months, and four months, respectively.

Various treatment intervals were selected to evaluate the efficacy of different treatment regimens in minimizing infection prevalence. The outcome of the study showed that dogs in Group A revealed a lower prevalence of infection compared to those in Group B and Group C. In addition, a higher prevalence of infection was seen in stray dogs compared to owned dogs in all groups.

However, certain limitations were observed in this study. The treatment of owned dogs was fully dependent on the compliance of the owners for administration of drugs appropriately. In ideal set up drug administration ought to be done by a veterinarian or assigned healthcare professional to ensure correct dosage and consistent submission. In such a set up the result of obtaining an even lower prevalence of *Echinococcosus granulosus* infection was possible. In the context of stray dogs, it is necessary to consider other contributing factors that resulted in higher prevalence of infection. Even though the population of dogs are comparable in the three villages, certain variation still existed such as the duration of dog's exposure to infectious sources can influence difference in the infection rate.

Overall, the same interval administration of dog deworming measures in all three villages was impeded due to less accessibility to some parts of the villages. In addition, the financial cost of assigning a health professional to administer the medication to each household was challenging. These combined factors compromise the effectiveness of the intervention in Ifrane region of Morocco. In addition, the re-infection rate is partly determined by seasonal variation. Seasonal variations can also impact the density of vegetation covering an area. Seasons also determined the survival of parasitic eggs in the environment. Moderate temperature and humidity allow it to last in its infectious state longer. Autumn/ early winter data was not stated to differentiate discrepancies since the study lasted only for 4 to 8 months (3).

Seasonal variability was found to be a key determinant factor in a study conducted in Ganze Tibetan region China. The result highlighted the importance of increasing awareness of re-infection for better preventive measures in future action plans. In this particular study possible factors that are known to be influential on dog chemotherapy including duration of treatment, weight of the dog, age and sex remained insignificant as opposed to varying weather conditions in different seasons on the outcome of echinococcus infection burden (46).

In the context of dog deworming a further study was performed in different provinces of middle Atlas Morocco that include cestocidal treatment of dogs coupled with sheep vaccination with EG95 vaccination. The sites are considered to have numerous numbers of dogs. The sheep

vaccination was given in two scenarios where in Ain Leuh 2 both treatment regimens of dogs and sheep was administered and in the remaining cities Ain Leuh 1, El Kbab and Ait Ishaq only sheep vaccination was given with their respective control group. A four-year follow-up of sheep was done using ultrasound and postmortem necropsy of infection detecting viable and non-viable cysts. The control group of sheep in each site was found to have high prevalence 88% in Ain Leuh ,40% in Al Ishaq and 46% in El kbab. In the treatment group, 600 dogs received praziquantel treatment. Prior to treatment the prevalence was 35% and 12% post treatment. Though this is a significant reduction of rate of re-infection, the annual incidence rate is still remarkably high with risk of 4% per month. This is due to the high environmental burden of infection and the absence of preventive measures. Particular interest to Ain Leuh 2 both treatment of dogs with praziquantel and vaccination of sheep was implemented. Based on the results of ultrasound finding of viable cysts, chemotherapy of dogs did not significantly contribute to the effect of sheep vaccination. This could possibly be due to contamination of the environment by stray dogs that were not included in the treatment interventions (47).

A different approach taken in pastoral region of tibettan regions of China where dog treatment was coupled with strict control of street and unleashed dogs was applied for a period of average life span of the adult worm. This was done through enforcing leash laws and undergo registration of dogs, resulting in a marked reduction of free dog roaming behavior of 93.2% of leashed dogs throughout the 24 hours in two villages. This increased the effectiveness of dog management strategies. Slight variation in the remaining villages (7.1%) in Rizha, (16%) in Eduoma for night roaming of dogs was recorded. From practical point of view in Morocco this was not possible due to inadequate infrastructure to monitor compliance and the important role of dogs in daily human activities (45).

3.2 Lack of surveillance and biosecurity measures

As part of understanding of the extent and magnitude of *echinococcus* transmission in endemic regions, determining the prevalence of *Echinococcus granulosus* infection in canine population is highly important. Adequacy of infrastructure in seven slaughterhouses were evaluated in Sid kacem region of Morocco which includes assessing the presence or absence of the necessary equipment (septic tank, incineration tank) and set up (room division, fence) for hygienic practices. Upon examination only 2 of the slaughterhouses had room divisions for the purpose of different

activities slaughter, skinning, hair removal, visceral removal, and cleaning. Similarly, only one slaughterhouse had an enclosed fence. Septic tanks and incineration tanks were found in two of the slaughterhouses, but none of the slaughterhouses fully met most of the necessities. Similar techniques of the slaughtering process were put in application in all slaughterhouses. Due to lack of partitions or separate areas, subsequent stages of slaughtering process of skinning and hair removal were performed in the same room. The lack of partitions or separate rooms also made the process of meat inspection challenging. The veterinary technicians only inspect the meat that is prepared to be taken to the local market. Since the area is overly occupied and overcrowded it makes conducting inspection inaccurate. The inspectors also stated that the butchers deliberately remove cysts from the meat that will be inspected which makes the inspection process results to be doubtful.

There is a certain pattern of discarding the "not sellable" parts of the animal, where the visceral parts will be collected and thrown away to a municipal garbage bin or taken to the local market to intentionally provide it to the free roaming dogs. The inspection activities are primarily done on the meat that will be sold on the market. Most of the time inspection of visceral organs (liver and lungs) are overlooked which is the contributing factor for the disease to go unnoticed (56).

Some of the veterinary technicians were hesitant to do a thorough inspection which is associated to multiple factors. One significant factor is the lack of cooperation observed by the inhabitants. In addition, constant resistance is encountered from slaughterhouse owners and butchers. Such resistance arises from the backing and support they get from the local political authorities. The lack of understanding about the magnitude of the disease within powerful stakeholders contributed to the existing problem. There is a wrong association of strict surveillance measures with decreased animal productivity and economical gain limitations obtained from meat supply chain and sale of animal parts. This in turn led to suppressing the voices of veterinary health professionals who engaged in reforms to improve the safety of slaughterhouses (4).

Another study conducted in Sid kacem showed that dogs in rural communities have shown an overall higher prevalence rate more so than dogs living in urban communities with a prevalence of 38.0% and 18.8% respectively. This is more evident in dogs that are living in close proximity of rural slaughterhouses with a prevalence of 62.7% (95% CI 48.1–75.9%) than dogs living in communities with no slaughterhouse in close vicinity (29.1%, 95% CI 21.7–37.2%) (48).

3.3 Limited awareness and understanding of *cystic echinococcosis*

It was reported in a case study that assessed the knowledge of *Echinococcus granulosus* infection in the same region where from the 543 persons surveyed inhabitants 50% of them have no familiarity with the infection. In addition, those that have some degree of awareness about the disease, only 21.3 % have a clear understanding dog's impact in the transmission of the disease. These inhabitants stated that the lack of street side waste collection by responsible authorities has facilitated easy access of free roaming dogs. Interviews conducted with the butchers revealed that the majority is aware of the disease but the presence of free roaming dogs around slaughterhouses is considered beneficial. This is because such dogs indirectly take part in the waste disposal by consuming the infected or otherwise visceral organs cleaning the surrounding environment.

Focus group discussion was done with selected inhabitants of middle Atlas Morocco. They believed discarding the carcasses of animals was found to be necessary to prevent access to dogs. This was done by burying it underground most of the time, throwing it in the river and burning the offal. However, the health professionals are skeptical about the effectiveness of burying the offal since it is not deep enough dogs can dig and take it out with ease. Even with these methods in place, proper dog food is believed to be expensive in the region so feeding the visceral organs to the owned dogs is still believed to be a good substitute feed (49).

Sufficient work was not done in stakeholder engagement inhibiting integrated public health programs for disease surveillance, monitoring and evaluation. Inadequate efforts were observed especially with higher authorities in engaging the farmers in facilitating knowledge sharing and collaboration. This resulted in resistance and limited readiness to cooperate in certain treatment programs. This was seen in the region of Ain Leuh 1, El kbab and Aiq Ishaq. These villages are in higher mountains of Morocco and known to have largest herd size estimated to be 240,000 and 65,000 animals in Aïn Leuh and El Kbab/Ait Ishaq regions, respectively. It is necessary to allocate resources and support so as to involve the breeders in hydatid disease controlling activities (47).

The complex nature of the *cystic echinococcosis* masked the extent of the infection in different provinces of Morocco. It also caused a wide knowledge gap on the disease dynamics and

underestimation of the prevalence of *cystic echinococcosis* among different stakeholders. In rural villages of Ait Ichou, Zaouiate Ait Isshak, El Kbab and Tighssaline variety of discussions and knowledge sharing meeting underwent and it showed that different levels of disease perception were observed. Health professionals such as doctors and nurses believe the disease has reduced, citing a reduction in limited patients with *cystic echinococcosis.* The local abattoirs also mentioned they observe fewer numbers of cattle and sheep with hydatid cysts in slaughterhouses. Health professionals did not put into consideration that the disease stays asymptomatic for years, so patients are less likely to visit clinics unless at a late stage. They also mentioned that the cattle and sheep that are brought for slaughtering are of young age which are too young to harbor the parasite. This influenced their perception of a reduction in the prevalence of the disease within their community (50).

This was also evidently seen in Sidi Kacem Province (North-West of Morocco), where 10 local abattoirs were selected for evaluation over the period of four consecutive years (April 2009–March 2013). Breeders brought ruminants predominantly of young age for slaughtering more commonly than older animals. On the same note a prevalence of 11.0% in sheep, 1.5% in goats and 42.9% in cattle were observed in Sid Karem province. Animals <3 years of age were brought for slaughtering. The study showed that there is a strong connection between increased rate of infection and increasing age of animals. This indicates that older animals that had exposure to egg infested grazing environment for an extended period of time continue to become possible sources of infection. The presence of such discrepancies and misconceptions reflects lack of communication among the responsible sectors hindering collaborative efforts. As a result, most of the control interventions stay suboptimal, not reaching their respective goal (49).

4. Discussion

The choice of Morocco as a primary focus of interest for reasons of high prevalence of infection in this country and the increasingly larger number of human hydatid disease cases. Regardless of the initiation of national hydatic control program in 2005, the country faced significant amount of socioeconomic loss associated with the disease (51).

Several countries obtained different outcomes as a result of their implemented hydatid control programs. Epidemiological state, economical factor, surveillance activities and health related

infrastructure in every country determine the likelihood of success or failure of the control measures in different countries. Iceland was one of the countries with substantial burden of human *cystic echinococcosis* infection. One fifth of the population was infected. Application of the hydatid control programs that incorporated a one health strategy was applied. New Zealand also implemented similar control measures that included veterinary interventions to reduce the transmission rate, campaigns that advocate hygiene practices and surveillance of slaughterhouses which led to complete eradication in both countries in 2002. Though health education has greatly contributed to the success of these programs, low human population, a significant number of inhabitants being literate, short slaughtering season have all led to a positive implication in the implementation of the programs. In addition, animals that are of young age became the preferred choice for slaughtering, typically lambs of 5 months of age which are too young to harbor viable cysts .Furthermore, geographical set up of Iceland, which is an isolated country with restricted end points permits easier control of moving stray dogs and border control unlike Morocco that has a dispersed geographical set up with a lot of less accessible remote areas (41).

As part of hydatid control program, praziquantel, a standard anthelmintic drug is administered to dogs 8 times per year to cease the parasite from getting into the gravid stage and preventing release of eggs thereby limiting environmental contamination. Effective implementation in the administration of the drug relies on proper infrastructure to reach out to the endemic areas regularly throughout the year. This was the case observed in South America once considered a model country, where praziquantel treatment in combination of insular models was implemented for over 30 years targeting complete eradication. However, the status of present hydatid control in the country is less than suboptimal (52).

Effective administration of dog chemotherapy becomes more challenging in rural areas where animal husbandry is a common practice. This is because owned dogs mostly assist in activities such as livestock herding. This leaves the dogs unattended, increasing the possibility of reinfection. This was taken into consideration by Tibetan communities, China. Emphasis was given to improving “roaming behavior” of owned dogs by leashing and tethering for a significant period throughout the day. In addition, “changing feeding habits,” reducing the number of stray dogs were applied in combination with regular deworming of their dogs (8 times a year). Such combined strategies allowed restriction of contact of dogs with livestock improving the effectiveness of the treatment (45).

So far in Morocco the implementation of cestodal treatment has a limited success. This is partly due to the exclusion of stray dogs in the program. Due to the large numbers of stray dogs in many regions of the country, the infection persists. In middle Atlas regions of Morocco, the prevalence of echinococcosis infection in livestock animals ranges from 51.3% to 68.5%. In addition, the large number of canid wild species has served as definitive hosts for disease transmission. Involvement of all dogs in medical management would allow rapid decline in the number of years of hydatid control programs presumed to achieve control of the disease (47).

Livestock such as sheep and cattle are the intermediate hosts in the transmission dynamics of *Echinococcosus granulosis* infection. The density of *Echinoccosis granulosus* eggs in the grazing environment determines the infection rate in these animals. EG95 vaccine developed in recent years has emerged as integrated control measure to jointly reduce the prevalence of *cystic echinoccosis* infection. The vaccine was implemented in many countries where the transmission is between dogs and sheep population. In Rio Negro province of Argentina vaccination of lambs was implemented in combination with dog deworming four times per year and lasted for eight years. Most of the lambs obtained two doses initially and a later booster dose. During each round of vaccination, the immunization coverage accounts for an average of 85%. Effectiveness of vaccine was assessed through necropsy examination and determination of number of cysts per sheep. The result showed marked reduction in the prevalence from before and after the introduction of the vaccine with 56.3% & 21.1%. respectively (53).

Similarly, a combined control strategies that includes vaccination of sheep with EG95 along with a quarterly base deworming of dogs was applied in mountainous region of Morocco. A 97% of immune protection rate were observed demonstrated by ultrasound and necropsy results. However, dogs deworming with praziquantel have not shown significant reduction in the incidence of sheep infection. This is due to deworming being done in owned dogs and did not include stray dogs which provided inadequate coverage to minimize the density of parasitic eggs in the environment. The dispersed population density in this region and limited infrastructure also created challenges to reach out a scheduled treatment in a timely manner (47). Based on the nature of the disease and from the experience seen with isolated praziquantel treatment of dogs for many years in several countries, the impact of sheep vaccination will be highly efficient and will be able to confine the infection transmission rate if implemented correctly (53).

Extended health education campaigns that include various sectors in a community require collaborative efforts and active engagement of different sectors to achieve behavioral change related to *cystic echinococcosis*. Evaluating the level of knowledge about the mechanism of the disease, mode of transmission and preventive measures among inhabitants in each area is part of the integral management of *cystic echinococcosis*. Such assessment was done in endemic region of Pakistan where 400 people constituting dog owners and butchers. The results revealed that only 4.1% are aware of the disease, 20% of the inhabitants are engaged in home slaughtering and consumption of uncooked meat and organs (liver and lung). 7.61% of the butchers are aware of the disease, however they stated that all the stray dogs consume the visceral organs nearby the slaughterhouses (54).

The middle Atlas region of Morocco is known for its pastoral communities. Few attentions have been given so far to reaching out to the community with health education outreach programs. This was evidenced through a survey done in this region. Home slaughtering of sheep and cattle is a commonly practiced tradition in this region. Due to improper disposal practices owned and stray dogs can freely access the offal. In addition, very few of the inhabitants had a complete knowledge of the disease and its impact indicating more work is needed from responsible sectors to improve the effectiveness of hydatid control programs (5).

5. Conclusion

Cystic echinococcosis is a neglected zoonotic disease reported by WHO (World health organization) as one of the 20 neglected zoonotic diseases. It is predominantly impacted low- and middle-income countries like Morocco. “Lack of resources” does add negative value in the spread of the disease. It limits dog treatment coverage, usage of advanced diagnostics and treatments such as ELISA (Enzyme-Linked Immunosorbent Assay), sheep vaccination, respectively. It also hinders training programs for health professionals and conducting further research in the regions of Morocco that are mostly affected by hydatid disease (47).

However, it is also important to note that like most other common zoonotic infectious diseases that occur in this country such as rabies, brucellosis and leishmaniasis, the underlying factors to most unsuccessful control measures are beyond “lack of resources “and are found to be multifactorial. These include improper resource allocation, lack of commitment from responsible sectors, competing interest that arise in setting a priority to the disease and lack of sufficient awareness

about the relationship between human and animal health has worsened the magnitude of the disease in Morocco (50).

It is essential to prepare guidelines adapted to the national health care system and depend on the disease prevalence in the country. It should include a clear set of definitions that determine what level of reduction in sheep and human infections is considered significant (55).

Species variability in the intermediate host contributes to the complexity of the parasite's lifecycle, distribution pattern of the disease and host-parasite interactions. Strategies that mitigate *cystic echinococcosis* transmission should put into consideration such variability.

The effectiveness of different treatment modalities in both hosts is co-dependent on health education. Health education should aim at interrupting the intermediate/ definitive host cycle. It should also bring about behavioral change ranging from community level to the higher authorities. Ideally, treatment of dogs with praziquantel eight times a year is the mainstay in the control of *cystic echinococcosis*. From a practical point of view this measure can be challenging in regions such as Northern Europe currently facing difficulty with infested foxes. It is also difficult to implement this modality of treatment to stray dogs due to ecological and logistical factors. Strict regulations and registration of dog population, regulated slaughterhouses, thorough inspection of meat and avoiding home slaughtering determine the effectiveness of the treatment.

Incorporation of biosecurity measures should be part of every control program. Intensive measures such as isolation of animals that are infected prior implementation of control program enables the reduction in disease prevalence at a faster rate. In resource limited countries such as Morocco maldistribution of resources makes the *cystic echinococcosis* endemic parts of the country to remain in a steady state (6).

References

1.Nunnari, Giuseppe, Marilia R. Pinzone, Salvatore Gruttadauria, Benedetto M. Celesia, Giordano Madeddu, Giulia Malaguarnera, Piero Pavone, Alessandro Cappellani, and Bruno Cacopardo. 'Hepatic Echinococcosis: Clinical and Therapeutic Aspects'. *World Journal of Gastroenterology* 18, no. 13 (7 April 2012): 1448–58. https://doi.org/10.3748/wjg.v18.i13.1448.

2.Azlaf, Rkia, and Allal Dakkak. 'Epidemiological Study of the Cystic Echinococcosis in Morocco'. *Veterinary Parasitology* 137, no. 1 (15 April 2006): 83–93. https://doi.org/10.1016/j.vetpar.2006.01.003.

3.Amarir, Fatima Ezzahra, Aouatif Saadi, Tanguy Marcotty, Abdelkbir Rhalem, Mohamed Oukessou, Hamid Sahibi, Majdouline Obtel, Mohammed Bouslikhane, Abderrahim Sadak, and Nathalie Kirschvink. 'Cystic Echinococcosis in Three Locations in the Middle Atlas, Morocco: Estimation of the Infection Rate in the Dog Reservoir'. *Vector Borne and Zoonotic Diseases (Larchmont, N.Y.)* 20, no. 6 (June 2020): 436–43. https://doi.org/10.1089/vbz.2019.2538.

4.Chebli, Houda, Abderrhamane Laamrani El Idrissi, Mustapha Benazzouz, Badre Eddine Lmimouni, Haddou Nhammi, Mourad Elabandouni, Mohammed Youbi, et al. 'Human Cystic Echinococcosis in Morocco: Ultrasound Screening in the Mid Atlas through an Italian-Moroccan Partnership'. *PLoS Neglected Tropical Diseases* 11, no. 3 (March 2017): e0005384. https://doi.org/10.1371/journal.pntd.0005384.

5.Thys, Séverine, Hamid Sahibi, Sarah Gabriël, Tarik Rahali, Pierre Lefèvre, Abdelkbir Rhalem, Tanguy Marcotty, Marleen Boelaert, and Pierre Dorny. 'Community Perception and Knowledge of Cystic Echinococcosis in the High Atlas Mountains, Morocco'. *BMC Public Health* 19, no. 1 (28 January 2019): 118. https://doi.org/10.1186/s12889-018-6372-y.

6. ‘Echinococcosis’. Accessed 13 August 2023. https://www.who.int/news-room/fact-sheets/detail/echinococcosis.

7.McManus, Donald P., Wenbao Zhang, Jun Li, and Paul B. Bartley. ‘Echinococcosis’. *Lancet (London, England)* 362, no. 9392 (18 October 2003): 1295–1304. https://doi.org/10.1016/S0140-6736(03)14573-4.

8.Davidson, Rebecca K., Thomas Romig, Emily Jenkins, Morten Tryland, and Lucy J. Robertson. ‘The Impact of Globalisation on the Distribution of Echinococcus Multilocularis’. *Trends in Parasitology* 28, no. 6 (June 2012): 239–47. https://doi.org/10.1016/j.pt.2012.03.004.

9.Romig, Thomas. ‘Epidemiology of Echinococcosis’. *Langenbeck’s Archives of Surgery* 388, no. 4 (September 2003): 209–17. https://doi.org/10.1007/s00423-003-0413-3.

10. Eckert, J., Gemmell, M.A., Meslin, F.X., & Pawłowski, Z.S. (2001). WHO/OIE manual on echinococcosis in humans and animals: a public health problem of global concern.

11.WOAH - World Organisation for Animal Health. ‘Echinococcosis’. Accessed 13 August 2023. https://www.woah.org/en/disease/echinococcosis/.

12.Moro, Pedro L. ‘Epidemiology and Control of Echinococcosis - UpToDate’, 3 May 2023. https://www.uptodate.com/contents/epidemiology-and-control-of-echinococcosis.

13.Kern, Peter. ‘Echinococcus Granulosus Infection: Clinical Presentation, Medical Treatment and Outcome’. *Langenbeck’s Archives of Surgery* 388, no. 6 (December 2003): 413–20. https://doi.org/10.1007/s00423-003-0418-y.

14.Silva, António Menezes da. ‘Human Echinococcosis: A Neglected Disease’. *Gastroenterology Research and Practice* 2010 (2010): 583297. https://doi.org/10.1155/2010/583297.

15.Prevention, CDC-Centers for Disease Control and. ‘CDC - Echinococcosis - Biology’, 16 July 2019. https://www.cdc.gov/parasites/echinococcosis/biology.html.

16.Casulli, Adriano, Alessandro Massolo, Urmas Saarma, Gérald Umhang, Federica Santolamazza, and Azzurra Santoro. ‘Species and Genotypes Belonging to Echinococcus Granulosus Sensu Lato Complex Causing Human Cystic Echinococcosis in Europe (2000-2021): A Systematic Review’. *Parasites & Vectors* 15, no. 1 (28 March 2022): 109. https://doi.org/10.1186/s13071-022-05197-8.

17.Zarrabi Ahrabi, S., R. Madani, B. Shemshadi, Sh Ranjbar Bahadori, and H. Hashemzadeh Farhang. 'Genetic Affinity of Echinococcus Granulosus Protoscolex in Human and Sheep in East Azerbaijan, Iran'. *Archives of Razi Institute* 75, no. 1 (March 2020): 47–54. https://doi.org/10.22092/ari.2018.122733.1227.

18.Dinkel, Anke, Kirsten Tackmann, Christoph Schulze, Peter Kutzer, Wilhelm Drewes, Thomas Romig, and Ute Mackenstedt. *First Record of the Pig Strain (G7) of Echinococcus Granulosus in Germany*, 2006. https://doi.org/10.13140/RG.2.2.28698.34240.

19.Thompson, R. C. A., A. C. Boxell, B. J. Ralston, C. C. Constantine, R. P. Hobbs, T. Shury, and M. E. Olson. 'Molecular and Morphological Characterization of Echinococcus in Cervids from North America'. *Parasitology* 132, no. 3 (March 2006): 439–47. https://doi.org/10.1017/S0031182005009170.

20.Villalobos, N., L. M. González, J. Morales, A. S. de Aluja, M. I. Jiménez, M. A. Blanco, L. J. S. Harrison, R. M. E. Parkhouse, and T. Gárate. 'Molecular Identification of Echinococcus Granulosus Genotypes (G1 and G7) Isolated from Pigs in Mexico'. *Veterinary Parasitology* 147, no. 1–2 (20 June 2007): 185–89. https://doi.org/10.1016/j.vetpar.2007.03.021

21.Deplazes, P., L. Rinaldi, C. A. Alvarez Rojas, P. R. Torgerson, M. F. Harandi, T. Romig, D. Antolova, et al. 'Global Distribution of Alveolar and Cystic Echinococcosis'. *Advances in Parasitology* 95 (2017): 315–493. https://doi.org/10.1016/bs.apar.2016.11.001.

22.Sanchez, Leny, Holger Mayta, Luis M. Jara, Manuela Verástegui, Robert H. Gilman, Luis A. Gómez-Puerta, and Cesar M. Gavidia. 'Echinococcus Granulosus Sensu Stricto and E. Canadensis Are Distributed in Livestock of Highly Endemic Area in the Peruvian Highlands'. *Acta Tropica* 225 (January 2022): 106178. https://doi.org/10.1016/j.actatropica.2021.106178.

23.Chaâbane-Banaoues, Raja, Myriam Oudni-M'rad, Jacques Cabaret, Selim M'rad, Habib Mezhoud, and Hamouda Babba. 'Infection of Dogs with Echinococcus Granulosus: Causes and Consequences in an Hyperendemic Area'. *Parasites & Vectors* 8 (17 April 2015): 231. https://doi.org/10.1186/s13071-015-0832-3.

24.Torgerson, P. R., and D. D. Heath. 'Transmission Dynamics and Control Options for Echinococcus Granulosus'. *Parasitology* 127 Suppl (2003): S143-158. https://doi.org/10.1017/s0031182003003810.

25.Bardonnet, K., M. C. Benchikh-Elfegoun, J. M. Bart, S. Harraga, N. Hannache, S. Haddad, H. Dumon, D. A. Vuitton, and R. Piarroux. 'Cystic Echinococcosis in Algeria: Cattle Act as Reservoirs of a Sheep Strain and May Contribute to Human Contamination'. *Veterinary Parasitology* 116, no. 1 (29 August 2003): 35–44. https://doi.org/10.1016/s0304-4017(03)00255-3.

26.Hernández, Felipe, Claudio Verdugo, Francisca Cárdenas, Rodrigo Sandoval, Nelly Morales, Pablo Olmedo, Andrea Bahamonde, Dennis Aldridge, and Gerardo Acosta-Jamett. 'Echinococcus Granulosus in the Endangered Patagonian Huemul (Hippocamelus Bisulcus)'. *Journal of Wildlife Diseases* 55, no. 3 (July 2019): 694–98.

27.Mohamed, M. Ibrahem, M. Ibrahem Wafa, M. Ibrahem Kawther, and B. Annajar Badereddin. 'Incidence and the History of Echinococcus Granulosus Infection in Dogs within the Past Few Decades in Libya: A Review'. *Journal of Veterinary Medicine and Animal Health* 9, no. 1 (31 January 2017): 1–10. https://doi.org/10.5897/JVMAH2016.0525.

28.Agudelo Higuita, Nelson Iván, Enrico Brunetti, and Cindy McCloskey. 'Cystic Echinococcosis'. *Journal of Clinical Microbiology* 54, no. 3 (March 2016): 518–23. https://doi.org/10.1128/JCM.02420-15.

29.Otero-Abad, Belen, and Paul R. Torgerson. 'A Systematic Review of the Epidemiology of Echinococcosis in Domestic and Wild Animals'. *PLoS Neglected Tropical Diseases* 7, no. 6 (2013): e2249. https://doi.org/10.1371/journal.pntd.0002249.

30.Zhang, Wenbao, Shengyue Wang, and Donald P. McManus. 'Echinococcus Granulosus Genomics: A New Dawn for Improved Diagnosis, Treatment, and Control of Echinococcosis'. *Parasite (Paris, France)* 21 (2014): 66. https://doi.org/10.1051/parasite/2014066.

31.Kinkar, Liina, Teivi Laurimae, Sami Simsek, Ibrahim Balkaya, Adriano Casulli, Maria Teresa Manfredi, Francisco Ponce-Gordo, et al. 'High-Resolution Phylogeography of Zoonotic Tapeworm Echinococcus Granulosus Sensu Stricto Genotype G1 with an Emphasis on Its Distribution in Turkey, Italy and Spain'. *Parasitology* 143, no. 13 (November 2016): 1790–1801. https://doi.org/10.1017/S0031182016001530.

32.Laurimäe, Teivi, Liina Kinkar, Vanessa Andresiuk, Karen Luisa Haag, Francisco Ponce-Gordo, Gerardo Acosta-Jamett, Teresa Garate, Luis Miguel Gonzàlez, and Urmas Saarma. 'Genetic Diversity and Phylogeography of Highly Zoonotic Echinococcus Granulosus

Genotype G1 in the Americas (Argentina, Brazil, Chile and Mexico) Based on 8279bp of MtDNA'. *Infection, Genetics and Evolution: Journal of Molecular Epidemiology and Evolutionary Genetics in Infectious Diseases* 45 (November 2016): 290–96. https://doi.org/10.1016/j.meegid.2016.09.015.

33.Brunetti, Enrico, Peter Kern, Dominique Angèle Vuitton, and Writing Panel for the WHO-IWGE. 'Expert Consensus for the Diagnosis and Treatment of Cystic and Alveolar Echinococcosis in Humans'. *Acta Tropica* 114, no. 1 (April 2010): 1–16. https://doi.org/10.1016/j.actatropica.2009.11.001.

34.Carmena, D., L. P. Sánchez-Serrano, and I. Barbero-Martínez. 'Echinococcus Granulosus Infection in Spain'. *Zoonoses and Public Health* 55, no. 3 (April 2008): 156–65. https://doi.org/10.1111/j.1863-2378.2007.01100.x.

35.'Echinococcosis - Annual Epidemiological Report for 2020', 23 May 2022. https://www.ecdc.europa.eu/en/publications-data/echinococcosis-annual-epidemiological-report-2020.

36.Karshima, Solomon Ngutor, Musa Isiyaku Ahmed, Nuhu Bala Adamu, Abdullahi Alhaji Magaji, Musa Zakariah, and Konto Mohammed. 'Africa-Wide Meta-Analysis on the Prevalence and Distribution of Human Cystic Echinococcosis and Canine Echinococcus Granulosus Infections'. *Parasites & Vectors* 15 (5 October 2022): 357. https://doi.org/10.1186/s13071-022-05474-6.

37.Azlaf, Rkia, Allal Dakkak, Abdelaziz Chentoufi, and Mokhtar El Berrahmani. 'Modelling the Transmission of Echinococcus Granulosus in Dogs in the Northwest and in the Southwest of Morocco'. *Veterinary Parasitology* 145, no. 3–4 (30 April 2007): 297–303. https://doi.org/10.1016/j.vetpar.2006.12.014.

38.Budke, Christine M., Peter Deplazes, and Paul R. Torgerson. 'Global Socioeconomic Impact of Cystic Echinococcosis'. *Emerging Infectious Diseases* 12, no. 2 (February 2006): 296–303. https://doi.org/10.3201/eid1202.050499.

39.Lightowlers, M. W., S. B. Lawrence, C. G. Gauci, J. Young, M. J. Ralston, D. Maas, and D. D. Heath. 'Vaccination against Hydatidosis Using a Defined Recombinant Antigen'. *Parasite Immunology* 18, no. 9 (September 1996): 457–62. https://doi.org/10.1111/j.1365-3024.1996.tb01029.x.

40.Labanchi, José Luis, Thelma V. Poggio, Ariana Gutiérrez, Guillermo Mujica, Daniel Araya, Claudia Grizmado, Arnoldo Calabro, et al. 'Analysis of Vaccination Strategy against Cystic Echinococcosis Developed in the Province of Río Negro, Argentina: 12 Years of Work'. *Veterinary Parasitology* 310 (October 2022): 109790. https://doi.org/10.1016/j.vetpar.2022.109790.

41.Craig, P. S., and E. Larrieu. 'Control of Cystic Echinococcosis/Hydatidosis: 1863-2002'. *Advances in Parasitology* 61 (2006): 443–508. https://doi.org/10.1016/S0065-308X(05)61011-1.

42.Larrieu, Edmundo, and Fabian Zanini. 'Critical Analysis of Cystic Echinococcosis Control Programs and Praziquantel Use in South America, 1974-2010'. *Revista Panamericana De Salud Publica = Pan American Journal of Public Health* 31, no. 1 (January 2012): 81–87. https://doi.org/10.1590/s1020-49892012000100012.

43.Harriott, Lana, Matthew Gentle, Rebecca Traub, Rowland Cobbold, and Ricardo Soares Magalhães. 'Geographical Distribution and Risk Factors for Echinococcus Granulosus Infection in Peri-Urban Wild Dog Populations'. *International Journal for Parasitology. Parasites and Wildlife* 10 (December 2019): 149–55. https://doi.org/10.1016/j.ijppaw.2019.08.005.

44.Yang, Yu Rong, Donald P. McManus, Yan Huang, and David D. Heath. 'Echinococcus Granulosus Infection and Options for Control of Cystic Echinococcosis in Tibetan Communities of Western Sichuan Province, China'. *PLoS Neglected Tropical Diseases* 3, no. 4 (2009): e426. https://doi.org/10.1371/journal.pntd.0000426.

45.Weng, Xiaodong, Zhiqiang Mu, Xu Wei, Xu Wang, Qingqiu Zuo, Shuo Ma, Youzhong Ding, et al. 'The Effects of Dog Management on Echinococcus Spp. Prevalence in Villages on the Eastern Tibetan Plateau, China'. *Parasites & Vectors* 13 (21 April 2020): 207. https://doi.org/10.1186/s13071-020-04082-6.

46.Wang, Qian, Wen-Jie Yu, Bo Zhong, Jing-Ye Shang, Liang Huang, Alexander Mastin, null Renqingpengcuo, et al. 'Seasonal Pattern of Echinococcus Re-Infection in Owned Dogs in Tibetan Communities of Sichuan, China and Its Implications for Control'. *Infectious Diseases of Poverty* 5, no. 1 (5 July 2016): 60. https://doi.org/10.1186/s40249-016-0155-4.

47.Amarir, Fatimaezzahra, Abdelkbir Rhalem, Abderrahim Sadak, Marianne Raes, Mohamed Oukessou, Aouatif Saadi, Mohammed Bouslikhane, et al. 'Control of Cystic Echinococcosis

in the Middle Atlas, Morocco: Field Evaluation of the EG95 Vaccine in Sheep and Cesticide Treatment in Dogs'. *PLoS Neglected Tropical Diseases* 15, no. 3 (March 2021): e0009253. https://doi.org/10.1371/journal.pntd.0009253.

48. Dakkak, A., I. El Berbri, A. F. Petavy, F. Boué, M. Bouslikhane, O. Fassi Fihri, S. Welburn, and M. J. Ducrotoy. 'Echinococcus Granulosus Infection in Dogs in Sidi Kacem Province (North-West Morocco)'. *Acta Tropica*, The Fate of Neglected Zoonotic Diseases, 165 (1 January 2017): 26–32. https://doi.org/10.1016/j.actatropica.2016.07.007.

49. El Berbri, Ikhlass, Marie J. Ducrotoy, Anne-Françoise Petavy, Ouaffa Fassifihri, Alexandra P. Shaw, Mohammed Bouslikhane, Franck Boue, Susan C. Welburn, and Allal Dakkak. 'Knowledge, Attitudes and Practices with Regard to the Presence, Transmission, Impact, and Control of Cystic Echinococcosis in Sidi Kacem Province, Morocco'. *Infectious Diseases of Poverty* 4 (9 November 2015): 48. https://doi.org/10.1186/s40249-015-0082-9.

50. Saadi, Aouatif, Nicolas Antoine-Moussiaux, Hamid Sahibi, Hind Filali, Tanguy Marcotty, and Séverine Thys. 'Feasibility and Acceptability of a New Integrated Approach to Control Cystic Echinococcosis in Morocco: Vaccination of Sheep and Anthelmintic Treatment of Dogs'. *Preventive Veterinary Medicine* 192 (July 2021): 105373. https://doi.org/10.1016/j.prevetmed.2021.105373.

51. 'The Socio-Economic Burden of Cystic Echinococcosis in Morocco: A Combination of Estimation Method | PLOS Neglected Tropical Diseases'. Accessed 13 August 2023. https://journals.plos.org/plosntds/article?id=10.1371/journal.pntd.0008410.

52. Larrieu, Edmundo, and Fabian Zanini. 'Critical Analysis of Cystic Echinococcosis Control Programs and Praziquantel Use in South America, 1974-2010'. *Revista Panamericana De Salud Publica = Pan American Journal of Public Health* 31, no. 1 (January 2012): 81–87. https://doi.org/10.1590/s1020-49892012000100012.

53. Larrieu, Edmundo, Guillermo Mujica, Daniel Araya, Jose Luis Labanchi, Marcos Arezo, Eduardo Herrero, Graciela Santillán, et al. 'Pilot Field Trial of the EG95 Vaccine against Ovine Cystic Echinococcosis in Rio Negro, Argentina: 8 Years of Work'. *Acta Tropica* 191 (March 2019): 1–7. https://doi.org/10.1016/j.actatropica.2018.12.025.

54. Khan, Aisha, Kashf Naz, Haroon Ahmed, Sami Simsek, Muhammad Sohail Afzal, Waseem Haider, Sheikh Saeed Ahmad, Sumaira Farrakh, Wu Weiping, and Guan Yayi. 'Knowledge, Attitudes and Practices Related to Cystic Echinococcosis Endemicity in Pakistan'.

Infectious Diseases of Poverty 7, no. 1 (22 January 2018): 4. https://doi.org/10.1186/s40249-017-0383-2.

55.Craig, Philip S., Donald P. McManus, Marshall W. Lightowlers, Jose A. Chabalgoity, Hector H. Garcia, Cesar M. Gavidia, Robert H. Gilman, et al. 'Prevention and Control of Cystic Echinococcosis'. *The Lancet. Infectious Diseases* 7, no. 6 (June 2007): 385–94. https://doi.org/10.1016/S1473-3099(07)70134-2.

56.Bardosh, Kevin Louis, Ikhlass El Berbri, Marie Ducrotoy, Mohammed Bouslikhane, Fassi Fihri Ouafaa, and Susan C. Welburn. 'ZOONOTIC ENCOUNTERS AT THE SLAUGHTERHOUSE: PATHWAYS AND POSSIBILITIES FOR THE CONTROL OF CYSTIC ECHINOCOCCOSIS IN NORTHERN MOROCCO'. *Journal of Biosocial Science* 48 Suppl 1 (September 2016): S92–115. https://doi.org/10.1017/S0021932015000486.

57.Cohen, H., E. Paolillo, R. Bonifacino, B. Botta, L. Parada, P. Cabrera, K. Snowden, et al. 'Human Cystic Echinococcosis in a Uruguayan Community: A Sonographic, Serologic, and Epidemiologic Study'. The American Journal of Tropical Medicine and Hygiene 59, no. 4 (October 1998): 620–27. https://doi.org/10.4269/ajtmh.1998.59.620.

58.Hodžić, Adnan, Amer Alić, Jovana Šupić, Vedad Škapur, and Georg Gerhard Duscher. 'Echinococcus Ortleppi, the Cattle Strain in a Crested Porcupine (Hystrix Cristata): A New Host Record'. Veterinary Parasitology 256 (30 May 2018): 32–34. https://doi.org/10.1016/j.vetpar.2018.05.004