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# Epidemiology, Molecular, and Phylogenetic Characterization of *Echinococcus granulosus* Cysts in Slaughtered Farm Animals in Al-Jouf Province, Saudi Arabia

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**ABSTRACT**

Echinococcosis, or hydatidosis, is a serious veterinary disease and public health issue worldwide, particularly in rural areas in which dogs have frequent contact with local herbivores. This study assessed the frequency of hydatidosis found among farm animals slaughtered in Al-Jouf Province in northern Saudi Arabia in 2021. A total of 156754 sheep, 36337 goats, 8590 camels, and 986 cattle were inspected for hydatidosis infection by comprehensive evaluation involving meticulous visual inspection and manual exploration of the internal organs through palpation. The cysts were subjected to molecular and phylogenetic analysis. The overall prevalence rates of hydatid cysts were 0.43%, 0.19%, 0.54%, and 0.51% in the inspected sheep, goats, camels, and cattle, respectively. The highest disease prevalence rates among sheep (27.8%) and goats (30.9%) occurred in the spring, and the highest prevalence rates among camels (41.3%) and cattle (80%) were in the summer. Regarding the prevalence of the disease in four slaughterhouses in the Al-Jouf Province, the highest prevalence in sheep, goats, and camels was in the Tabarjal slaughterhouse (1.43%, 0.81%, and 1.08%, respectively), although the Al-Qurayat slaughterhouse had the highest prevalence rate in cattle (1.98%). Complete molecular analysis indicated that cytochrome c oxidase subunit 1 (*cox1*) sequences from cyst isolates belonged to *Echinococcus granulosus* (*E. granulosus*). Moreover, there was high homology (98-100%) with associated Genbank sequences of *E. granulosus* isolated from sheep in the Kingdom of Saudi Arabia (KSA). Sheep and camels were a significant source of hydatidosis transmission to dogs and helped to maintain disease incidence in the Al-Jouf Province. Thus, significant efforts should focus on preventing cyst transmission from abattoirs and infected stray dogs.

**Keywords:** *Echinococcus granulosus*, Epidemiology, Molecular characterization, Farm animals

## INTRODUCTION

Echinococcosis, or hydatidosis, is a parasitic disease caused by infection with a larva from the *Echinococcus* genus of the family Taeniidae (WHO, 2016). It is a widespread zoonotic disease affecting several areas of the world, including Saudi Arabia, posing a serious risk to humans and livestock and having a severe economic impact on the farm industry (Eckert and Deplazes, 2004; WHO, 2015). There are multiple species of *Echinococcus (E.)*, including *E. granulosus*, *E. multilocularis, E. oligarthrus,* and *E. vogeli*, of which *E. granulosus* is most often associated with disease (Nabavi et al., 2014).

The *E. granulosus* lifecycle requires both an intermediate and a definitive host. Intermediate hosts include herbivores such as sheep and cattle, while the final host is a definitive carnivore (Eckert et al., 2001; Eslami et al., 2016). Humans are aberrant dead-end hosts who become infected by contacting the feces of definitive hosts or after eating food contaminated by parasite eggs (Virginio et al., 2012). Since canines and domestic animals have the most contact in rural areas, domestic animals serve as the primary reservoir for human disease (Almalki et al., 2017).

In Saudi Arabia, the major meat-producing livestock includes sheep, goats, camels, and cattle. At a total of 13,444,435 heads, sheep are the primary source of meat (72%), with a high number being imported to fulfill the needs of the Saudi population (GASTAT, 2018). Echinococcosis affects both humans and their domestic animals in several countries in Saudi Arabia, mainly in the Western Region (Hayajneh et al., 2014).

Infection can lead to financial losses from the disposal of diseased organs and reduced milk, meat, and wool production (Singh et al., 2016). Moreover, helminth infections not only hinder productivity but also have detrimental effects on food quality (Toni et al., 2023). Infected animals can also be potential sources of co-contamination that can impact humans and other animals (Singh et al., 2016).

Since Echinococcosis is a significant zoonotic disease, it is critical to assess the prevalence and incidence of infection in potential intermediate hosts. This defines the role of each animal species in parasite conservation and disease movement (Cadavid Restrepo et al., 2016). Accurate disease diagnosis is required for the detection of the livestock source responsible for transmission (Cadavid Restrepo et al., 2016). In intermediate hosts, hydatidosis is usually



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asymptomatic and thus often overlooked by farmers (Elham et al., 2014). Inspection of meat from slaughtered animals is important for identifying cysts and determining disease prevalence. As a result, slaughterhouses remain the ideal location to study hydatidosis in livestock from different regions (Almalki et al., 2017). Numerous research studies have addressed the prevalence of Echinococcosis disease in Saudi Arabia. For instance, a study by Merst et al. (2023) explored the infection rates among stray dogs and slaughtered animals in the Al-Kharj region in Saudi Arabia, revealing a 14% infection rate in dogs and rates of 9.18%, 7.5%, and 10% for sheep, goats, and camels, respectively. Another study in the eastern region of Saudi Arabia identified three *Echinococcus* species in camels *E. granulosus*, *E. Canadensis*, and *E. ortleppi* (Al-Hizab et al., 2021). The prevalence of Echinococcosis, a zoonotic disease, is notable among livestock in the Arabian Peninsula countries (Al-Shaibani et al., 2021) and is a global concern (Alvi and Alsayeqh., 2022).

A deeper understanding of *Echinococcus* species will inform new methods of control, diagnosis, and treatment (McManus, 2010). Sequencing of the whole mitochondrial genome has proved extremely useful to the phylogenetic analyses of species of *Echinococcus* (Nakao et al., 2013). The current study aimed to determine the presence of *E. granulosus* over one year among sheep, goats, camels, and cattle and to assess the impact of season, region, and *Echinococcus* species on disease prevalence in Al-Jouf Province, Saudi Arabia. Hydatid cysts were evaluated by polymerase chain reaction (PCR), and DNA specific for the mitochondrial *cox1* gene was sequenced to compare with the genotype *E. granulosus* in slaughtered livestock in Saudi Arabia.

## MATERIALS AND METHODS

### Ethics approval

This study was approved by the Research Ethical Committee of Jouf University in Saudi Arabia (approval No: 40-08-48)

### Study location

The study was conducted using slaughtered goats, sheep, camels, and cattle from four slaughterhouses in the Al-Jouf province located in the north of Saudi Arabia between longitudes (36° and 41°) east and latitudes (28° and 32°) north from January to December 2021. All slaughterhouses were operating in accordance with the Technical Regulations for Meat Inspection issued by the Ministry of Municipal and Rural Affairs in 2008.

### Study procedure

A total of 156,754 slaughtered sheep, 36,337 goats, 8,590 camels, and 986 cattle were examined by veterinarians for cystic hydatidosis (Table 1). Region, season, and species information was obtained, and each slaughtered animal was comprehensively evaluated through meticulous visual inspection and manual exploration of the internal organs through palpation, according to Eckert et al. (1984). Ten hydatid protoscolices from each animal species (random selection) were dipped in Phosphate Buffer Saline PBS and transferred to the parasitology laboratory, Biology department, college of Science in Jouf University into sterile test tubes (each sample originating from a single cyst), fixed in 70% ethanol, and stored at -20°C for DNA extraction.

**Table 1.** Number and infection rate (%) of hydatidosis in slaughterhouses in the Al-Jouf Province, Saudi Arabia in 2021

| Slaughterhouse location | Sheep | Goats | Camels | Cattle | Total |
|---|---|---|---|---|---|
| **Sakaka** | 131/91620 (0.14%) | 11/19830 (0.06%) | 14/4673 (0.3%) | 1/580 (0.17) | 157/116703 (0.14%) |
| **Dumat al-Jandal** | 39/16572 (0.24%) | 7/3222 (0.22%) | 7/1322 (0.53%) | 0/161 (0%) | 53/21277 (0.25%) |
| **Al-Qurayat** | 349/37400 (0.93%) | 33/11188 (0.3%) | 16/1763 (0.91%) | 4/202 (1.98%) | 402/50553 (0.80%) |
| **Tabarjal** | 160/11162 (1.43%) | 17/2097 (0.81%) | 9/832 (1.08%) | 0/43 (0%) | 186/14134 (1.32%) |
| **Total** | 679/156754 (0.43%) | 68/36337 (0.19%) | 46/8590 (0.54%) | 5/986 (0.51%) | 798/202667 (0.39%) |
| **P value** | 0.207 | 0.186 | 0.042* | 0.334 | 0.295 |

Each box of this table includes information on the number of infected animals/number of animals examined and Infection rates (%). * $p < 0.05$ (significant)


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**Examination of cysts and viability of protoscoleces**

The cyst fluid was extracted using a needle and subsequently examined under a light microscope to determine the presence of protoscoleces (Eckert et al., 1984).

**DNA extraction**

DNA was extracted using the DNeasy Blood and Tissue Kit (Qiagen, Germany) according to the manufacturer's instructions. Hydatid cyst samples (25 mg) were processed, and DNA was dissolved in 50 µl elution buffer.

**PCR amplification**

According to a previous study by Bowles et al. (1992), the mitochondrial *cox1* gene was targeted with the forward primer 5'- TTTTTTGGGCATCCTGAGGTTTAT-3' and the reverse primer

5'-TAAAGAAAGAACATAATGAAAATG-3' (Vivantis Technologies Sdn. Bhd, Malaysia) in a 25 µl total reaction volume with 12.5 µl of COSMO-PCR-RED Master Mix (W1020300X, Willofort, UK), 0.5 µL (17 µM) of each primer, and 2 µL of target DNA (Bowles et al., 1992). The PCR program involved 5 minutes of denaturation at 95°C, 40 cycles of denaturation at 95°C for 20 seconds, annealing at 55°C for 30 seconds, extension at 72°C for 45 seconds, and a single extension step at 72°C for 10 minutes. The PCR products (450 bp) were analyzed using 1.2% agarose gel electrophoresis with a 100 bp Plus DNA ladder and analyzed using an InGenius3 gel documentation system (Syngene, UK).

**Phylogenetic tree construction**

Three cytochrome c oxidase subunit I (*cox1)* sequences from human isolates involved in this study (EEG1-EEG3) were registered in GenBank under the accession numbers MZ348904-MZ348906, and three nucleotides from camel isolates (EEG4-EEG6) were registered under the accession numbers, MZ348907-MZ348909. Positive PCR products for the *cox1* gene were sequenced (Macrogen, Korea) and analyzed using BioEdit 7.0.4 and MUSCLE. The resultant sequences were aligned with the *E. granulosus*. *cox1* reference sequences using a neighbor-joining method of the aligned sequences in the CLC Sequence Viewer 6 program, UK.

**Statistical analysis**

The findings were presented in the form of tables using descriptive analysis. The data obtained from the study were subjected to statistical analysis using SPSS software version 26. To assess the significance of the presence of hydatidosis in various locations and animal species, the Chi-square ($X^2$) test was employed, and the results were presented in Table 1. Additionally, the uniform distribution test was utilized to evaluate the significance of hydatidosis occurrence across different seasons and animal species, with the corresponding outcomes documented in Table 2. A p-value less than 0.05 shows a significant difference.

**Table 2.** Seasonal prevalence of hydatidosis in slaughtered animals in the Al-Jouf Province, Saudi Arabia in 2021

| Season \ Animals | Sheep No. (%) | Goats No. (%) | Camels No. (%) | Cattle No. (%) |
|---|---|---|---|---|
| **Winter** | 173 (25.5) | 15 (22.1) | 11 (23.9) | 1 (20) |
| **Spring** | 189 (27.8) | 21 (30.9) | 9 (19.6) | 0 (0) |
| **Summer** | 169 (24.9) | 15 (22.1) | 19 (41.3) | 4 (80) |
| **Autumn** | 148 (21.8) | 17 (25) | 7 (15.2) | 0 (0) |
| **Total** | 679 (100) | 68 (100) | 46 (100) | 5 (100) |
| **P-value** | 0.524 | 0.687 | 0.048* | 0.305 |

* $p < 0.05$ (significant), No: Number.

## RESULTS

An overall hydatidosis prevalence of 0.43%, 0.19%, 0.54%, and 0.39% was detected among sheep, goats, camels, and cattle, respectively, obtained from four slaughterhouses in Al-Jouf Province, for a total prevalence of 0.39% (Table 1).

Of the four slaughterhouses in the Al-Jouf Province (Table 1), the Tabarjal slaughterhouse had the highest prevalence of hydatidosis in sheep, goats, and camels (1.43%, 0.81%, and 1.08%, respectively), while the Al-Qurayat slaughterhouse had the highest prevalence in cattle (1.98%). The Dumat al-Jandal slaughterhouse had the lowest prevalence of sheep (0.24%), the Sakaka slaughterhouse had the lowest in goats (0.06%) and camels (0.3%), and the Dumat al-Jandal and Tabarjal slaughterhouses had the lowest for cattle (0%). Statistically, the difference was not significant ($p > 0.05$) for all animals of the four slaughterhouses, barring for the camels, which had a significant difference ($p < 0.05$), as shown in Table 1.



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The highest prevalence of disease occurred in spring for sheep and goats (27.8% and 30.9%, respectively) and in summer for camels and cattle (41.3% and 80%, respectively), while the lowest prevalence occurred in autumn for sheep and camels (21.8% and 15.2%, respectively), in winter and summer for goats (22.1%), and in spring and autumn for cattle (0%, Table 2). Statistically, the difference was not significant ($p > 0.05$) for all animals of the four Seasons, except camels, which had a significant difference ($p < 0.05$), as shown in Table 2.

A 450 bp fragment was amplified from all cyst samples using *cox1* PCR (Figure 1). A phylogenetic tree of the isolates showed that they belonged to *E. granulosus*. Whole isolate sequences of the mtDNA revealed 98–100% homology with the *E. granulosus* reference sequence, MZ350810, isolated from sheep in Al-Taif, KSA, and MN720282 isolated from sheep in Al-Madinah, KSA (Figure 2).

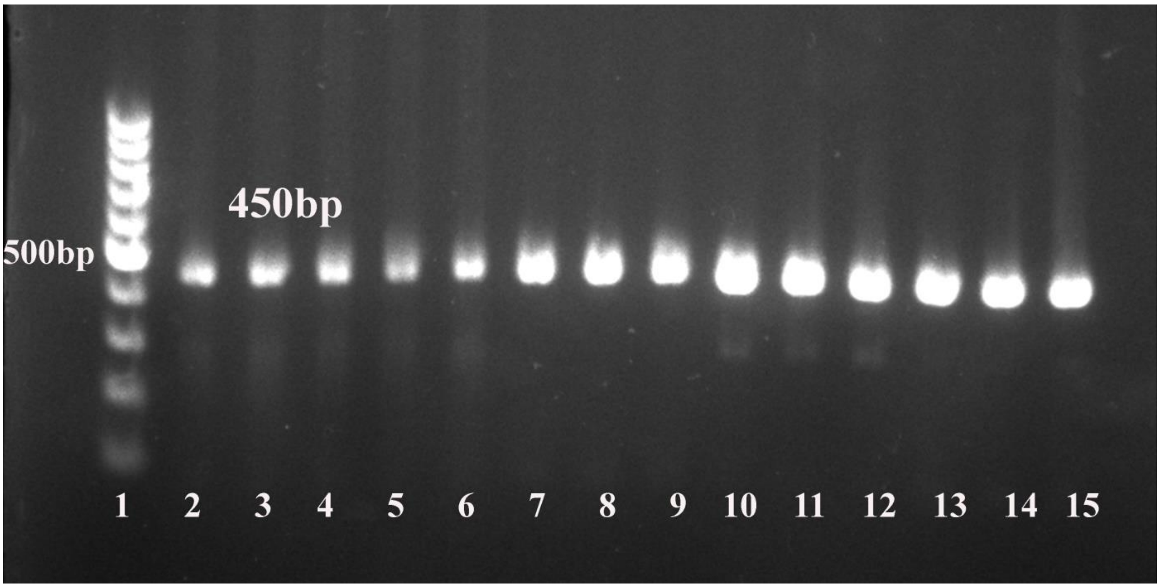


**Figure 1.** PCR analysis of the *cox1* gene for *Echinococcus granulosus* revealed a 450 bp band derived from representative sheep (lanes 2-5), goats (lanes 6-9), camels (Lanes 10-12), and cattle (Lanes 13-15)

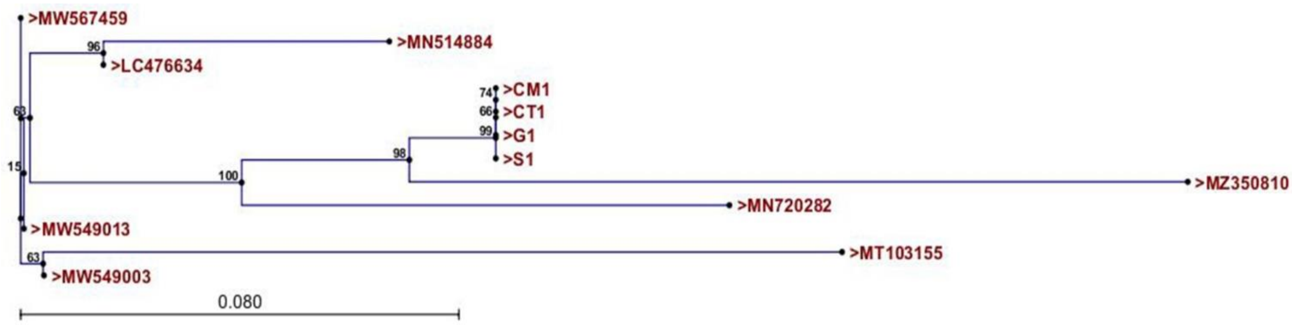


**Figure 2.** Phylogenetic tree of *Echinococcus granulosus* sequences isolated from sheep (S1), goats (G1), camels (CM1) and cattle (CT1) in Egypt and reference sequences from other *Echinococcus* species using the neighbor-joining method based on the *cox1* gene

## DISCUSSION

Hydatid disease is one of the most transmissible and virulent zoonotic helminth infections worldwide. The disease has had a major health and economic impact on several countries (Raissi et al., 2021). In Saudi Arabia, hydatidosis is of particular concern to sheep farmers (Eckert et al., 2001; Ibrahim, 2010; Toulah et al., 2012; Almalki et al., 2017; Al-Shaibani et al., 2021; Al Malki and Ahmed, 2022), leading to great financial losses resulting from the disposal of diseased organs and a decline in meat production (Singh et al., 2014). Diseased animals also serve as possible sources of transmission to dogs, humans, and other animals (Singh et al., 2014). There is limited information on the rate of *E. granulosus* infection in farm animals in Saudi Arabia. Surveys of abattoirs are a vital data source on the prevalence of hydatidosis in different livestock since reliable disease diagnosis occurs using meat inspection (El-Ghareeb et al., 2017; Almalki et al., 2017). The reported Hydatid disease prevalence ranges from 0.4 to 41.6% in camels, 0.0 to 40.5% in cattle, 0.5 to 30.5% in goats, and 0.1 to 69.6% in sheep (Almalki et al., 2017). Most research has been conducted in the


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western and central regions of the country. The current study is the first to evaluate the prevalence of hydatidosis in sheep, goats, camel, and cattle meat from slaughterhouses in Al-Jouf, a province located in Northern Saudi Arabia. Although many studies indicate the spread of the Hydatid disease in other regions of the Kingdom of Saudi Arabia (Haroun et al., 2008; Ibrahim, 2010; Toulah et al., 2012; Fdaladdin et al., 2013; Hayajneh et al., 2014; Almalki et al., 2017; El-Ghareeb et al., 2017; Al-Hizab et al., 2018; Merst et al., 2023).

A total of 156,754 slaughtered sheep, 36,337 goats, 8,590 camels, and 986 cattle were inspected for cystic hydatidosis from January 2021 to December 2021. The prevalence of the disease was 0.43%, 0.54%, 0.39%, and 0.19% among the inspected sheep, camels, cattle, and goats, respectively, with a collective prevalence of 0.39%. A prior study found that the prevalence rate of Echinococcosis was 0.60%, 0.51%, 0.47%, and 0.38% in cattle, camels, goats, and sheep, respectively, with a total prevalence of 0.42% (Toulah et al., 2017). Unlike the current study, in which the highest disease prevalence was observed in camels, the highest prevalence rate was found in cattle, followed by camels, sheep, and goats. Former investigations conducted in Saudi Arabia and other countries have confirmed that sheep have a higher incidence of Echinococcosis than other animals, including goats and cattle (Ibrahim, 2010; Toulah et al., 2012; Hayajneh et al., 2014; Amer et al., 2018; Joanny et al., 2021).

In the current study, the incidence of cystic hydatidosis among sheep (0.43%) was the lowest among those reported in Saudi Arabia, including 1.06–2.33% in Riyadh (Almalki et al., 2017; Abdel-Baki et al., 2018), 12.61% in Al Baha (Ibrahim, 2010), 69.6% in Jeddah (Toulah et al., 2012), 9.6–13.5% in Al-Taif (Al-Malki and Degheidy, 2013; Hayajneh et al., 2014; Al Malki and Ahmed, 2022), 6.6.% in the Al-Ahsa region (El-Ghareeb et al., 2017), 9.6% in Al-Taif (Al-Malki and Degheidy, 2013), 6.8% in Najran (Almalki et al., 2017), 7.06% in Hail (Hasona et al., 2017) and 2.83% in Dammam (El-Ghareeb et al., 2017). There are several explanations for the differences in disease prevalence in different regions of Saudi Arabia, including different weather (rainfall and temperature) patterns, the timeframe in which the study was conducted, and the abundance of infected host dogs in each area (Hasona et al., 2017).

The prevalence of hydatidosis in camels (0.54%) was similar to that observed in Hail, Saudi Arabia (0.51%, Amer et al., 2018) but lower than that seen in Al Baha (37.5%, Ibrahim, 2010), Al-Madina Al-Munawwarah (34.6, Fdaladdin et al., 2013), Jeddah (41.6%, Toulah et al., 2017), and Egypt 5.4%, 7.4%, and 14.6% (Mousa et al., 2015; Dyab et al., 2018; Ahmed et al., 2021). The higher disease prevalence in camels than in other species may be related to several factors. First, camels may be infected with a strain that has adapted to be avirulent in cattle, goats, and sheep, or these other species may have developed immunity to strains that infect camels (Fdaladdin et al., 2013). Second, areas used to herd camels may have a high number of dogs, the final host responsible for transmitting the disease to camels (the intermediate hosts, Mirzaei and Fooladi, 2012). Third, the prevalence of hydatidosis may relate to camel herder-specific behaviors, including the way they raise and feed their camels and dogs (Mirzaei and Fooladi, 2012)**.**

The prevalence of hydatidosis in goats (0.19%) was similar to that observed in Makkah, in Saudi Arabia(0.23%, Alsulami, 2019) but lower than that seen in Al Baha (9.58%, Ibrahim, 2010), Al-Madina Al-Munawwarah (15.11%, Fdaladdin et al., 2013), Jeddah (30.56%, Toulah et al., 2017) and Iran (14.5%, Daryani et al., 2007). Meanwhile, the prevalence of hydatidosis in cattle (0.51%) was similar to that observed in Egypt (0.64%, El-Alfy et al., 2017) but lower than that seen in Al-Madina Al-Munawwarah (28.7%, Fdaladdin et al., 2013), Al Baha (9.35%, Ibrahim, 2010), Jeddah (31.13%, Toulah et al., 2017), and Hail (2.76%, Amer et al., 2018).

The current study found that sheep and camels had a higher prevalence of hydatidosis, and were thus more likely to be infected than cattle and goats. Thus, it is probable that the prevalence of cystic echinococcosis in northern Saudi Arabia is lower than it is in other regions of the country. However, controlling and treating stray dogs against worms, and cautiously removing polluted viscera continue to be vital to managing the problem (Amer et al., 2018).

The prevalence of hydatidosis in different animal species found at four slaughterhouses in the Al-Jouf Province in Saudi Arabia was highest in the Tabarjal slaughterhouse (1.32%), followed by Al-Qurayat (0.8%), Dumat al-Jandal (0.25%), and Sakaka (0.14%, Table 1). Differences in disease prevalence rates by location may be due to differences in the virulence of parasite strains found in each region as well as cultural and regional variances in dog behavior (McManus, 2006; Haridy et al., 2006; Hayajneh et al., 2014). The variation in infection rates among the four slaughterhouses studied can be attributed to the distinct characteristics of these regions, including their urban, semi-urban, or rural nature. Notably, the Tabarjal region, which is predominantly rural and hosts many farms where many affected dogs reside, recorded the highest infection rates. This finding aligns with previous studies by Almalki et al. (2017) and Abdel-Baki et al. (2018). Torgerson (2006) revealed that infection rates vary by country. This study also showed that infection rates were significantly higher than those identified in the same region during the prior year.

Hydatidosis infection rates changed considerably during the study period, with a higher prevalence in spring for sheep (27.8%) and goats (30.9%) and a higher prevalence in summer for camels (41.3%) and cattle (80%, Table 2). Prevalence infection rates then gradually decreased to their lowest point in autumn for sheep (21.8%) and camels (15.2%), in winter and summer for goats (22.1%), and in spring and autumn for cattle (0%).

The highest prevalence of hydatidosis observed in this study was among sheep in spring, and it was similar to that observed in Al Baha in Saudi Arabia (Ibrahim, 2010) but differed from that seen by Abdel-Baki et al. (2018) in winter,



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Amer et al. (2018) in summer, and Dyab et al. (2018) in autumn. The lowest prevalence of hydatidosis in autumn was similar to that observed by prior studies conducted in Saudi Arabia (Al-Qureishy, 2008) and Egypt (Dyab et al., 2018) but differed from studies conducted in the winter (Ibrahim, 2010), spring (Amer et al., 2018), and summer (Abdel-Baki et al., 2018).

The current study found that hydatidosis infection in goats was highest in the spring, whereas Toulah et al. found that rates were highest in summer (Toulah et al., 2017). The lowest prevalence of infection in goats was observed in winter and summer. This was similar to rates observed by Ibrahim (2010) but differed from other studies that observed the lowest prevalence in spring (Daryani et al., 2007) and autumn (El-Ghareeb et al., 2017) for goat's infection.

The current study found that hydatidosis infection in camels was highest in summer. This differed from other studies that found the highest infection rates in winter (Amer et al., 2018; Ahmed et al., 2021), spring (Ibrahim, 2010), and autumn (Dyab et al., 2018). The lowest prevalence of infection in camels was observed in autumn and was similar to rates reported by Amer et al. (2018) but differed from other studies that observed the lowest prevalence in winter (Toulah et al., 2017), spring (Ahmed et al., 2021), and summer (Ibrahim, 2010). Meanwhile, the current study found that infection in cattle was highest in summer; a similar observation is reported by Daryani et al. (2007). However, Amer et al. (2018) and El-Alfy et al. (2017) reported the highest rates in spring and autumn, respectively. The lowest prevalence of infection in cattle occurred in spring and autumn in the current study, and it was similar to that observed by Daryani et al. (2007).

Seasonal variations in infection may be due to variations in the age of livestock during different seasons (Ibrahim, 2010). Changes in climate variables, including temperature, moisture, and rainfall, also impact the disease spread (Elmajdoub and Rahman, 2015), as does the condition of pastures in different seasons and the density and accessibility of infected final host populations (Ernest et al., 2009; Ibrahim, 2010).

Mitochondrial DNA sequences (mtDNA) are extensively used for molecular categorization and phylogenetic analyses of *E. granulosus*. The *cox1* gene is the most frequently used mtDNA gene for phylogenetic analysis of helminth parasites (Mirbadie et al., 2019; Al Malki and Hussien, 2021). PCR and phylogenetic analysis revealed that among the livestock tested in this study, sheep were the primary host of *E. granulosus* in Saudi Arabia. This aligns with prior studies suggesting that sheep serve as the primary source of hydatidosis transmission (Abdel-Baki et al., 2018; Amer et al., 2018). Additionally, the present study revealed a high sequence similarity between the *cox1* gene of hydatid cyst isolates from sheep and the genotype of *E. granulosus* isolated from sheep in Al-Madinah, referencing sequence (MN720282) (Al-Mutairi et al., 2020).

## CONCLUSION

The findings highlight the need for a disease prevention plan to reduce *E. granulosus* infection in studied animals. Based on PCR and phylogenetic analysis of *E. granulosus* in the present study, more inclusive follow-up studies and surveys of abattoirs in different regions of Saudi Arabia are needed to verify the public health importance of hydatidosis. Additionally, it is recommended to improve monitoring of the slaughtering process and implement effective measures for managing stray dogs. Also, efforts should be intensified to control the transmission of cysts from slaughterhouses by properly disposing of the infected offal.

## DECLARATIONS

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### Availability of data and materials

The data of the current study are available.

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### Authors' contribution

Abdulsalam Alkhaldi collected and analyzed the data as well as wrote and revised the manuscript. Authors checked and confirmed the final draft of manuscript before submission to present journal.

### Ethical consideration

The author declares that this manuscript is original and is not being considered for publication elsewhere. Other ethical issues, including consent to publish, misconduct, fabrication of data, and redundancy, have been checked by the authors.

**Competing interests**

The author declared that they have no conflict of interest.

## REFERENCES


Abdel-Baki AS, Almalki E, and Al-Quarishy S (2018). Prevalence and characterization of hydatidosis in Najdi sheep slaughtered in Riyadh city, Saudi Arabia. Saudi Journal of Biological Sciences, 25: 1375-1379. DOI: https://www.doi.org/10.1016/j.sjbs.2018.04.011

Ahmed AB, Ras R, Mahmoud AF, El-Ghazaly E, Widmer G, Dahshan H, and Elsohaby I (2021). Prevalence and bacterial isolation from hydatid cysts in dromedary camels (*Camelus dromedarius*) slaughtered at Sharkia abattoirs, Egypt. Journal of Parasitic Diseases, 45: 236-243. DOI: https://www.doi.org/10.1007/s12639-020-01300-x

Al-Hizab FA, Hamouda MA, Amer OH, Edris AM, Ibrahim AM, Abdel-Raheem SM, and El-Ghareeb WR (2018). Hepatic cystic echinococcosis in camels of Saudi Arabia: Prevalence, risk factors and economic loss. Journal of Camel Practice and Research, 25: 295-302. DOI: https://www.doi.org/10.5958/2277-8934.2018.00041.3

Al-Hizab FA, Mohamed NS, Wassermann M, Hamouda MA, Ibrahim AM, El-Ghareeb WR, Abdel-Raheem SM, Romig T, and Omer RA (2021). Three species of *Echinococcus granulosus* sensu lato infect camels on the Arabian Peninsula. Parasitology Research, 120: 2077-2086. DOI: https://www.doi.org/10.1007/s00436-021-07156-1

Al Malki J and Ahmed N (2022). Epidemiological and histomorphic studies in sheep infected with hydatid cyst in Taif area. Saudi Journal of Biological Sciences, 29: 886-893. DOI: ,https://www.doi.org/10.1016/j.sjbs.2021.10.017

Al Malki JS and Hussien NA (2021). Molecular characterization and phylogenetic studies of *Echinococcus granulosus* and *Taenia multiceps* coenurus cysts in slaughtered sheep in Saudi Arabia. Open Life Sciences, 16: 1252-1260. DOI: https://www.doi.org/10.1515/biol-2021-0131

Almalki E, Al-Quarishy S, and Abdel-Baki AS (2017). Assessment of prevalence of hydatidosis in slaughtered Sawakny sheep in Riyadh city, Saudi Arabia. Saudi Journal of Biological Sciences, 24: 1534-1537. DOI: https://www.doi.org/10.1016/j.sjbs.2017.01.056

Al-Malki JS and Degheidy NS (2013). Epidemiological studies of hydatidosis among slaughtered sheep and humans in Taif, Saudi Arabia. Assiut Veterinary Medical Journal, 59(139): 45-50. DOI: https://www.doi.org/10.21608/avmj.2013.171910

Al-Mutairi, NM, Taha HA, and Nigm, AH (2020). Molecular characterization of *Echinococcus granulosus* in livestock of Al-Madinah (Saudi Arabia). Journal of Helminthol, 94: e157. https://www.doi.org/10.1017/S0022149X20000395

Al-Qureishy SA (2008). Prevalence of cestode parasites in sheep slaughtered in Riyadh City, Saudi Arabia. Journal of the Egyptian Society of Parasitology, 38: 273-280. Available at: https://europepmc.org/article/med/19143137

Al-Shaibani IRM, Al-Khadher AMA, and Al-Yahiri AGA (2021). Cystic echinococcosis among livestock in Arabia Peninsula: A systemic review and meta-analysis. Asian Journal of Research in Animal and Veterinary Sciences, 8: 183-195. Available at: https://sdiopr.s3.ap-south-1.amazonaws.com/doc/Revised-ms_AJRAVS_78491_v1.pdf

Alsulami M (2019). Prevalence and histopathological study on cystic hydatidosis in heart and spleen of goat slaughtered at Makkah, Saudi Arabia. Annals of Parasitology, 65(3): 225-236. DOI: https://www.doi.org/10.17420/ap6503.204

Alvi MA and Alsayeqh AF (2022). Food-borne zoonotic echinococcosis: A review with special focus on epidemiology. Frontiers in Veterinary Science, 9: 1072730. DOI: https://www.doi.org/10.3389/fvets.2022.1072730

Amer OH, Haouas N, Al-Hathal EAAR, El-Shikh I, and Ashankyty I (2018). Cystic echinococcosis in slaughtered animals in Ha'il, Northwestern Saudi Arabia. Japanese Journal of Veterinary Research, 66(4): 289-296. DOI: https://www.doi.org/10.14943/jjvr.66.4.289

Bowles J, Blair D, and McManus DP (1992). Genetic variants within the genus *Echinococcus* identified by mitochondrial DNA sequencing. Molecular and Biochemical Parasitology, 54(2): 165-173. DOI: https://www.doi.org/10.1016/0166-6851(92)90109-W

Cadavid Restrepo AM, Yang YR, McManus DP, Gray DJ, Giraudoux P, Barnes TS, Williams GM, Soares Magalhães RJ, Hamm NAS, and Clements ACA (2016). The landscape epidemiology of echinococcoses. Infectious Diseases of Poverty, 5: 13. DOI: https://www.doi.org/10.1186/s40249-016-0109-x

Daryani A, Alaei R, Arab R, Sharif M, Dehghan MH, and Ziaei H (2007). The prevalence, intensity and viability of hydatid cysts in slaughtered animals in the Ardabil Province of Northwest Iran. Journal of Helminthology, 81(1): 13-17. DOI: https://www.doi.org/10.1017/S0022149X0720731X

Dyab AK, Marghany ME, Othman RA, Ahmed MA, and Abd-Ella OH (2018). Hydatidosis of camels and sheep slaughtered in Aswan Governorate, Southern Egypt. Russian Journal of Parasitology, 12(3): 33-41. DOI: https://www.doi.org/10.31016/1998-8435-2018-12-3-33-41

Eckert J, Gemmell MA, Matyas Z, Soulsby EJ, and World Health Organization. (1984). Guidelines for surveillance, prevention and control of echinococcosis/hydatidosis (No. VPH/81.28). World Health Organization. https://policycommons.net/artifacts/515351/guidelines-for-surveillance-prevention-and-control-of-echinococcosishydatidosis-edited-by-j/1491469/

Eckert J and Deplazes P (2004). Biological, epidemiological, and clinical aspects of echinococcosis, a zoonosis of increasing concern. Clinical Microbiology Reviews, 17: 107-135. DOI: https://www.doi.org/10.1128/CMR.17.1.107-135.2004

Eckert J, Gemmell MA, Meslin FX, Pawlowski ZS, and World Health Organization (2001). WHO/OIE manual on echinococcosis in humans and animals: A public health problem of global concern. World Organization for Animal Health. Available at: https://apps.who.int/iris/handle/10665/42427

El-Alfy EN, Al-kappany YM, and Abuelwafa SA (2017). Parasitological and Pathological studies on Tissue parasites among slaughtered animals in Dakahlia Province, Egypt. Egyptian Veterinary Medical Society of Parasitology Journal, 13(1): 78-98. DOI: https://www.doi.org/10.21608/evmspj.2017.37734

El-Ghareeb WR, Edris AM, Alfifi AE, and Ibrahim AM (2017). Prevalence and histopathological studies on hydatidosis among sheep carcasses at Al-Ahsa, Saudi Arabia. Alexandria Journal of Veterinary Sciences, 55: 146-153. DOI: https://www.doi.org/10.5455/ajvs.285871

Elham M, Hassan B, Ghasem NA, Gholamreza R, and Parviz S (2014). Epidemiological study of hydatidosis in the dromedaries (*Camelus dromedarius*) of different regions of Iran. Asian Pacific Journal of Tropical Biomedicine, 4(Suppl. 1): S148-S151. DOI: https://www.doi.org/10.12980/APJTB.4.2014C725

Elmajdoub LO and Rahman WA (2015). Prevalence of hydatid cysts in slaughtered animals from different areas of Libya. Open Journal of Veterinary Medicine, 5(1): 53250. DOI: https://www.doi.org/10.4236/ojvm.2015.51001

Ernest E, Nonga HE, Kassuku AA, and Kazwala RR (2009). Hydatidosis of slaughtered animals in Ngorongoro district of Arusha region, Tanzania. Tropical Animal Health and Production, 41: 1179-1185. DOI: https://www.doi.org/10.1007/s11250-008-9298-z

Eslami A, Meshgi B, Jalousian F, Rahmani S, and Salari MA (2016). Genotype and phenotype of *Echinococcus granulosus* derived from wild sheep (*Ovis orientalis*) in Iran. Korean Journal of Parasitology, 54: 55-60. DOI: https://www.doi.org/10.3347/kjp.2016.54.1.55

Fdaladdin YAJ, Alsaggaf AI, and Wakid MH (2013). Comparative epidemiological studies on *echinococcosis* of local and imported livestock in Al-Madina Al-munawwarah in Saudi Arabia. Egyptian Journal of Hospital Medicine, 50: 108-126. DOI: https://www.doi.org/10.21608/ejhm.2018.16080

GASTAT (2019). Agriculture, water and environment. General authority for statistics. Statistical yearbook of 2018. Available at: https://www.stats.gov.sa/en/1011

Haridy FM, Ibrahim BB, Elshazly AM, Awad SE, Sultan DM, El-Sherbini GT, and Morsy TA (2006). Hydatidosis granulosus in Egyptian slaughtered animals in the years 2000-2005. Journal of the Egyptian Society of Parasitology, 36(3): 1087-1100. Available at: https://europepmc.org/article/med/17153715

Haroun EM, Omer OH, Mahmoud OM, and Draz A (2008). Serological studies on hydatidosis in camels in Saudi Arabia. Research Journal of Veterinary Sciences, 1(1): 71-73. Available at: https://scialert.net/abstract/?doi=rjvs.2008.71.73

Hasona NA, Amer OH, Morsi A, and Raef A (2017). Comparative biochemical, parasitological, and histopathological studies on cystic echinococcosis in infected sheep. Comparative Clinical Pathology, 26: 805-810. DOI: https://www.doi.org/10.1007/s00580-017-2450-2

Hayajneh FMF, Althomali AMH, and Nasr ATM (2014). Prevalence and characterization of hydatidosis in animals slaughtered at Al Taif abattoir, Kingdom of Saudi Arabia. Open Journal of Animal Sciences, 4(1): 38-41. DOI: https://www.doi.org/10.4236/ojas.2014.41006

Ibrahim MM (2010). Study of cystic echinococcosis in slaughtered animals in Al Baha region, Saudi Arabia: Interaction between some biotic and abiotic factors. Acta Tropica, 113(1): 26-33. DOI: https://www.doi.org/10.1016/j.actatropica.2009.08.029

Joanny G, Mehmood N, Dessì G, Tamponi C, Nonnis F, Hosri C, Saarma U, Varcasia A, and Scala A (2021). Cystic echinococcosis in sheep and goats of Lebanon. Parasitology, 148(7): 871-878. DOI: https://www.doi.org/10.1017/S0031182021000494

McManus DP (2006). Molecular discrimination of taeniid cestodes. Parasitology International, 55(Supplement): S31-S37. DOI: https://www.doi.org/10.1016/j.parint.2005.11.004

McManus DP (2010). Echinococcosis with particular reference to Southeast Asia. Advances in Parasitology, 72: 267-303. DOI: https://www.doi.org/10.1016/S0065-308X(10)72010-8

Mares MM, Abdel-Gaber R, and Al-Quraishy S (2023). Prevalence of *Echinococcus granulosus* (Cestoda: Taeniidae) infection in stray dogs and local herbivores in Al-Kharj, Saudi Arabia. Indian Journal of Animal Research, 57: 928-933. DOI: https://www.doi.org/10.18805/IJAR.BF-1553

Mirbadie SR, Najafi Nasab AN, Mohaghegh MA, Norouzi P, Mirzaii M, and Spotin A (2019). Molecular phylodiagnosis of *Echinococcus granulosus* sensu lato and *Taenia hydatigena* determined by mitochondrial *Cox1* and *SSU-rDNA* markers in Iranian dogs: Indicating the first record of pig strain (G7) in definitive host in the Middle East. Comparative Immunology, Microbiology and Infectious Diseases, 65: 88-95. DOI: https://www.doi.org/10.1016/j.cimid.2019.05.005

Mirzaei M and Fooladi M (2012). Prevalence of intestinal helminthes in owned dogs in Kerman city, Iran. Asian Pacific Journal of Tropical Medicine, 5(9): 735-737. DOI: https://www.doi.org/10.1016/S1995-7645(12)60116-3

Mousa WM, Mahdy OA, Abdel-Wahab AM, and El-Gameel OA (2015). Epidemiological and serological studies on *Cystic echinococcosis* among camels in Egypt. Journal of Parasitology - Photon Foundation, 105: 212-218. Available at: https://scholar.cu.edu.eg/?q=drolfat/files/photon.pdf

Nabavi R, Khedri J, and Saadati D (2014). Comparative prevalence of hepato-pulmonary hydatidosis among native and imported cattle in the north of Sistan and Baluchestan: Iran. Journal of Parasitic Diseases, 38: 371-373. DOI: https://www.doi.org/10.1007/s12639-013-0262-0

Nakao M, Lavikainen A, Yanagida T, and Ito A (2013). Phylogenetic systematics of the genus *Echinococcus* (*Cestoda: Taeniidae*). International Journal for Parasitology, 43(12-13): 1017-1029. DOI: https://www.doi.org/10.1016/j.ijpara.2013.06.002

Raissi V, Etemadi S, Sohrabi N, Raiesi O, Shahraki M, Salimi-Khorashad A, and Ibrahim A (2021). Molecular characterization and phylogeny of *Taenia hydatigena* and *Echinococcus granulosus* from Iranian sheep and cattle based on *COX1* gene. Current Microbiology, 78: 1202-1207. DOI: https://www.doi.org/10.1007/s00284-021-02377-0

Singh BB, Dhand NK, Ghatak S, and Gill JPS (2014). Economic losses due to cystic echinococcosis in India: Need for urgent action to control the disease. Preventive Veterinary Medicine, 113(1): 1-12. DOI: https://www.doi.org/10.1016/j.prevetmed.2013.09.007

Singh BB, Sharma R, Sharma JK, Mahajan V, and Gill JPS (2016). Histopathological changes associated with *E. granulosus* echinococcosis in food-producing animals in Punjab (India). Journal of Parasitic Diseases, 40: 997-1000. DOI: https://www.doi.org/10.1007/s12639-014-0622-4



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Toni NAD, Elmoneem SAA, Girgis JRA, Hussein AW, Thagfan FA, Abdel-gaber R, Ali SE, Marey AM, Al-najjar MAA, Alkhudhayri A et al. (2003). *In vitro* role of biosynthesized nanosilver from Allium sativum against helminths. Food Science and Technology, 43: e123622. DOI: ,https://www.doi.org/10.1590/fst.123622

Torgerson PR (2006). Mathematical models for the control of cystic echinococcosis. Parasitology International, 55 (Supplement): S253-S258. DOI: https://www.doi.org/10.1016/j.parint.2005.11.037

Toulah FH, El Shafei AA and Alsolami MN (2012). Prevalence of hydatidosis among slaughtered animals in Jeddah, Kingdom of Saudi Arabia. Journal of the Egyptian Society of Parasitology, 42: 563-572. DOI: https://www.doi.org/10.12816/0006341

Toulah FH, El Shafi AA, Alsolami MN and Wakid MH (2017). Hydatidosis among imported animals in Jeddah, Saudi Arabia. Journal of Liver and Clinical Research, 4(1): 1031. Available at: https://www.kau.edu.sa/Files/0030238/Researches/72635_45786.pdf

Virginio VG, Monteiro KM, Drumond F, de Carvalho MO, Vargas DM, Zaha A, and Ferreira HB (2012). Excretory/secretory products from *in vitro*-cultured *Echinococcus granulosus* protoscoleces. Molecular and Biochemical Parasitology,183: 15-22. DOI: https://www.doi.org/10.1016/j.molbiopara.2012.01.001

World Health Organization (WHO) (2015). Neglected tropical diseases. World Health Organization (2016). Echinococcosis fact sheet. Available at: https://www.who.int/news-room/fact-sheets/detail/echinococcosis/: *N 377*