

Incidence of Dicrocoeliasis Among Local and Imported Sheep at Riyadh Region

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Abstract. *Dicrocoelium dendriticum* is a common liver fluke of sheep and other herbivorous animals. Examination of sheep livers for *D. dendriticum* worms revealed infection rate reached to 22.3%. This rate was decreased to 14.86% ($P < 0.01$) after diagnoses of infection in the same animals via detection of eggs by fecal examination. The rate of infection considered to be low in local Naheemi sheep (6%) in comparison with the same sheep type imported from Turkey as the rate reached to 16.3%. There is an indirect relation sheep between the increase in the rate of infection and age of the examined sheep. The high rate of infection (52.9%) was recorded among sheep of 25-36 month old, followed by that of 13- 24 month old (18.4%) and the lowest rate of infection (9.6%) was recorded in sheep of age 6-12 month ($P < 0.01$) The seasonal distribution of dicrocoeliosis indicated a higher percentage of infection in winter and autumn (19.2% , 18.5% respectively) as compared with spring and summer (13.3% , 9.3% respectively). The infection with dicrocoeliasis only was 8% while dicrocoeliasis with other heminthic worms (*Fasciola gigantica*, *Moniezia expansa*, *Strongyloides papillosus*, *Trichuris globulosa*, *Nematodirus spathiger*, *Haemonchus contortus*, *Ostertagia circumcincta*, *Marshallagia marshalli*) was 6.86%. The study recommended application of other diagnostic methods rather than fecal examination during importation of living sheep suspected to be infected by a parasite like *Dicrocoelium* .

Introduction

Dicrocoeliosis *Dicrocoelium dendriticum* (Rudolphi, 1819; Loss, 1899) is a parasitic disease which mainly affects the liver of ruminants (Ferre *et al.*, 1994). *D. dendriticum* (the lancet fluke) has a worldwide distribution . It is commonly found in the bile ducts of sheep, goats, cattle, camels, rabbits, pigs, deer, horses, elk, dogs, and occasionally man (Nobles and Nobles, 1982) The infection is acquired from eating infected sheep liver. Dicrocoeliasis is now imposing itself as an animal and zoonotic helminthic disease in many Arab countries (Al-Mathal & Fouad, 2004) . The life cycle of the parasite is similar to that of other liver trematodes, however, its eggs are ingested by the first intermediate host which is a terrestrial snail of the genera *Cionella*, *Abida*, *Cochlicella*, *Helicella* and *Zebrina* (Rosicky and Groschaft, 1984). After being ingested as slime balls by a second intermediate host of ants comprising any of 14 species of genus *Formica*. The parasite develops in these ants to infective metacercariae that are ingested by the definitive mammals hosts to develop into adults in the bile duct (Srivastava, 1975 and Wilson, 1991). The

clinical features range from asymptomatic or mild to moderately severe or even fatal disease (Drabick *et al.*, 1988) . Dicrocoeliasis , known as " small liver fluke disease", is poorly known and often underestimated by researchers and practitioners in many countries (Otranto and Traversa, 2002) . The presence of *Dicrocoelium dendriticum* in humans faeces has been demonstrated in several archeo-parasitological surveys in the UK, in a site dated to 12th century (Taylor, 1955) and in France, in material excavated from a sealed latrine (dating from 11th to 16th centuries) (Bouchet, 1995). Recently, *Dicrocoelium* eggs have also been retrieved in Canada, in an archaeological site dated to 17th century (Horne and Tuck, 1996).

So, the aim of the present study is to determine the prevalence of dicrocoeliasis among local and imported sheep slaughtered at Riyadh, Saudi Arabia .

Material and Methods

During complete one year (beginning of May 2004 to end of April 2005), slaughtered sheep at main Riyadh abattoir, King Saudi Arabia, were inspected

before and after slaughtering concerning to infection by *D.dendriticum*.

Examined animals

A number of 50 local Naheemi and 300 Turkey imported sheep were investigated. The animals were identified by their ear's number, owner's name, and age pre slaughtering. Animals were allocated into three age groups as 6-12 month old, 13-24 month old and 25-36 month old. They still under observation using their ear number post slaughtering where their abdominal cavities were examined before evisceration.

Samples collected

Direct before slaughtering, rectal fecal sample was collect from each animal. Post slaughtering, the whole liver were collected from each target animal. The samples were identified, kept in ice box then transferred directly for examination in the laboratory

Examination of samples

Each sample was examined macroscopically for detection of whole worms or part of worm. The suspected trophozoies, large size eggs and larvae were diagnosed using direct smear method. Concentration flotation technique was adopted for diagnosis of *D.dendriticum* and other eggs according to (Solusby 1986). Two gram of fecal sample was mixed with suitable amount of concentrated salt solution. The mixture was sieved (mesh size 0.9mm) into two test tubs. The tubs were centrifuged at 1500 r.p.m. for 5 minutes. The tope surface layer of the supernatant solution was transferred into clean slide, cover slip was added then examine under the microscope. Formalin-ether method was adapted in some cases according to (Ash & Orihel 1991) .

Post mortum examination

The abdominal cavity of each slaughtered sheep was examined carefully by the necked eyes. Liver were directly separated from the carcuses. Livers were transferred directly to the laboratory, and directly examined using squeezing technique according to (Welch *et al*,1987). The livers were freshly sliced into 5-10 mm thickening slides. Directly immersed into worm water (37 °C) then strongly squeezed by both hands, living worms were migrate to worm water, the process were repeated 2-3 time where the water sieved and all the extracted worms or anterior part of worms were counted. The collected worms and different eggs were identified according to (Soulsby 1986).

Results

Results of the study are described in the illustrated five tables as the following:

Table 1. Incidence of infection by *D. dendriticum* among slaughtered sheep by detection of eggs in the feces and worms in livers .

No. examined	Eggs in feces		Worms in liver	
	-ve (%)	+ve (%)	-ve (%)	+ve (%)
350	298 (85.14)	52 (14.86)	272 (77.7)	78 (22.3)
Statistical analysis	$\chi^2 = 6.4$		$P < 0.01$	

Table 2. Rate of infection by *D. dendriticum* in local and imported Naheemi sheep.

Sheep source	Total No.	Rate of infection (eggs in feces)	
		-ve (%)	+ve (%)
Local Naheemi	50	47 (88)	3 (6)
Imported Naheemi (Turkey))	300	251 (83.7)	49 (16.3)
Statistical analysis	$\chi^2 = 3.6$		$P > 0.05$

Table 3. Relationship between infection rate with *D. dendriticum* and age of slaughtered sheep .

Age of sheep(month)	Total No.	Eggs in feces	
		-ve (%)	+ve (%)
6 – 12	208	188 (90.4)	20 (9.6)
13 - 24	125	102 (81.6)	23 (18.4)
25 - 36	17	8 (47.1)	9 (52.9)
Statistical analysis	$\chi^2 = 25.5$		$P < 0.01$

Table 4. Seasonal destreputation of dicrocoeliasis among the year

Year Seasons	No.exmined	-ve (%)	+ve (%)
winter	78	63 (80.8)	15 (19.2)
spring	83	72 (86.7)	11 (13.3)
Summer	97	88 (90.7)	9 (9.3)
autumn	92	75 (81.5)	17 (18.5)
Total No.	350	298 (85.1)	52 (14.9)

Discussion

Sheep are the most important livestock in Saudi Arabia and mutton is the preferred and most consumed meat throughout the Kingdom (Abu-Zinada, 2005) .

Dicrocoeliosis often remains clinically undetected or undiagnosed, most likely because of its subclinical nature (Ducommun and Pfister, 1991). Its diagnosis is mainly established by recovering adult worms in the liver at necropsy or detecting eggs at coprological examination. The later is the most commonly used technique for the diagnosis of dicrocoeliosis and can reveal the presence of small (40mm×25 mm), thick-walled, yellowishbrown eggs(Otranto and Traversa, 2002) .

Table 5. Relationship between infection with *D. dendriticum* and infection with other helminths among slaughtered sheep

Sheep source : (No.)		Eggs in feces			
		Total + ve (%)	Positive numbers between groups []		
			Only <i>Dicrocoelium</i> [A]	<i>Dicrocoelium</i> with other helminths [B]	Only other helminths [C]
(1)	Local Naheemi : (50)	21 (42)	1 (2)	2 (4)	18 (36)
(2)	Imported Naheemi (Turkey): (300)	108 (36)	27 (9)	22 (7.3)	59 (19.7)
(3)	Total : (350)	129 (36.8)	28 (8)	24 (6.86)	77 (22)
Statistical analysis	(1) & (2)	$\chi^2 = .67$ $P > 0.05$	$\chi^2 = 2.9$ $P > 0.05$	$\chi^2 = .73$ $P > 0.05$	$\chi^2 = 6.65$ $P < 0.05$
	(1) & (3)	$\chi^2 = .49$ $P > 0.05$	$\chi^2 = 2.3$ $P > 0.05$	$\chi^2 = .59$ $P > 0.05$	$\chi^2 = 4.67$ $P > 0.05$
	(2) & (3)	$\chi^2 = .053$ $P > 0.05$	$\chi^2 = .21$ $P > 0.05$	$\chi^2 = .064$ $P > 0.05$	$\chi^2 = .54$ $P > 0.05$

In the present study, post Mortum examination of slaughtered sheep livers (350) revealed infection by *Dicrocoelium* worms in 22.3%, while feces examination for eggs revealed low infection rate reached to 14.86% ($P < 0.01$), (Table 1). eggs in feces were small (35-45 x 22-30 μ m), embryonated, thick-shelled with dark brown colouration, The low rate of infection recorded after diagnosis of eggs in feces agreed with Rosicky and (Groschaft 1982) as they mentioned that *Dicrocoelium* eggs are not easily diagnosed in feces. Low rate of infection was recorded after examination of local Naheemi sheep showed 3 cases of infection with *D. dendriticum* (6%), In the same time high rate of infection with *D. dendriticum*, was recorded in the imported Naheemi sheep from Turkey showed 49 (16.3%) (Table 2). The seasonal distribution indicated a higher percentage of infection in winter and autumn (19.2%, 18.5% respectively) as compared with spring and summer (13.3%, 9.3% respectively) (Table 4). On the other hand, The high infection rate (52.9%) was recorded in sheep aged 25-36 month, followed by 18.4% in those aged 13-24, and the lowest was 9.6 among sheep aged 6-12 month ($P < 0.01$). Furthermore, dicrocoeliasis also was found in 8% in the sheep with mixed infection with other helminths (*Fasciola gigantica*, *Moniezia expansa*, *Strongyloides papillosus*, *Trichuris globulosa*, *Nematodirus spathiger*, *Haemonchus contortus*, *Ostertagia circumcincta*, *Marshallagia marshalli*). This was recorded in 6.86% in them (Table 5).

In Saudi Arabia, (Nasher 1990) recorded *D. dendriticum* in imported sheep in Asir province. The recorded incidence in the present study (16.3%) in the Turkey imported Naheemi sheep considered to be lower than that previously mentioned by (Gawish *et al.*, 1993) as they reported 28% infection rate at Riyadh region among condemned livers of slaughtered animals particularly Naheemi sheep imported from Turkey. In 1988, the incidence was 8.29% in Naheemi sheep

imported from Turkey, 0.274% in Nagdi (Central region of Saudi Arabia), 0.0924 in Merino sheep imported from Australia. In sheep slaughtered at Jeddah abattoirs, (Abu Zinada 1999), reported dicrocoeliasis by different percentage in imported and local breed sheep. The infection rate was 40% in sheep imported from Somalia, 26% in that from Turkey and it was 2% in native Saudi breed sheep. (Helmy and Al-Mathal 2003) reported dicrocoeliosis in 9-24% of sheep Slaughtered at Riyadh region. In Iraq, (Wajdi and Nassir (1983) reported *D. dendriticum* in the livers of sheep with 14.3%. In the present study, The highest rate of infection was recorded in winter season, this agreed with Manga-(Gonzalez *et al.* 1991) as they mentioned that in Spain. In Nigeria, (Nwosu and Srivastava 1993) recorded prevalence of *Dicrocoelium* egg excretion with 1.8% for sheep and goats. In India, 8.1% of sheep was positive for dicrocoeliosis with coprological studies and the necropsy liver examination of slaughtered sheep revealed 24.1% was infected with *D. dendriticum*. On other hand, The seasonal distribution of dicrocoeliosis indicated a higher percentage of infection in winter and autumn as compared with spring and summer (Jithendran and Bhat, 1996). Also, In Italy, recorded 67.5% of sheep examination was positive for *D. dendriticum* the survey show that *D. dendriticum* was widely and homogeneously spread throughout the study area southern Italian (Cringoli *et al.*, 2002).

The present study demonstrated high incidences for *Dicrocoelium* infection in the imported Naheemi sheep in comparison with the local breed of the same type. For this reason special interest must be focused during importation of sheep from Europe. Moreover avoid importation from the endemic localities considered to be one of the most important preventive measure for parasite like *Dicrocoelium* as diagnosis of infection via fecal examination considered as an easily matter as detailed describe in the present study.

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نسبة داء الديكروسلليم بين الغنم المحلية والمستوردة في منطقة الرياض

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الكلمات المفتاحية: أغنام، النعيمي، المحلية، المستوردة، الديكروسلليم، كبد، البراز.

ملخص البحث: الديكروسلليم من ديدان الكبد الشائعة بين الأغنام و آكلات الأعشاب. في هذه الدراسة تم تقدير الإصابة بهذا الطفيل على مدار عام في أغنام النعيمي المحلية والمستوردة من تركيا وذلك بتشخيص وجود البويضات الدالة على الطفيل في البراز ومقارنة ذلك

باستخلاص الديدان بعد فحص أكباد نفس الأغنام بعد ذبحها. وقد أوضحت الدراسة أن نسبة الإصابة التي تم تسجيلها بعد فحص الأكباد كانت أعلى (٢٢.٣٪) مقارنة بتلك التي تم الحصول عليها بعد تشخيص وجود بويضات الطفيل في البراز (١٤.٨٦٪). وقد أظهر الفحص

إنخفاض معدلات الإصابة في الأغنام المحلية (نعيمي) (٦٪) مقارنة بتلك المستوردة (نعيمي تركي) (١٦.٣٪). وأثبتت الدراسة وجود علاقة طردية بين التقدم في عمر الحيوان ومعدل الإصابة. حيث كانت معدلات الإصابة مرتفعة بين الأغنام التي تتراوح أعمارها من ٢٥-٣٦ شهراً (٥٢.٩٪) يليها عمر ١٣-٢٤ شهراً (١٨.٤٪) وأقلها في عمر ٦-١٢ شهراً (٩.٦٪). كذلك فقد أشار التوزيع الموسمي للإصابة بداء الديكروسلليم إلى إرتفاع معدل الإصابة في الشتاء والخريف (١٩.٢٪، ١٨.٥٪ على التوالي) مقارنة بالربيع والصيف (١٣.٣٪، ٩.٣٪ على التوالي). وقد تم تسجيل إصابة بديدان الديكروسلليم فقط في ٨٪ من الأغنام المصابة بينما اختلطت الإصابة مع وجود بويضات بعض الطفيليات الأخرى في نسبة بلغت ٦.٨٦٪ من الحيوانات التي تم فحصها. وقد شملت هذه الأنواع:

(*Fasciola gigantica*, *Moniezia expansa*, *Strongyloides papillosus*, *Trichuris globulosa*, *Nematodirus spathiger*, *Haemonchus contortus*, *Ostertagia circumcincta*, *Marshallagia marshalli*).

وقد خلصت الدراسة إلى أهمية الإعتماد على وسيلة أخرى غير فحص البراز عند استيراد الأغنام الحية من مناطق استيطان

الإصابة خاصة لطفيل مثل الديكروسلليم كما هو مبين تفصيلاً في هذا البحث.

