

## Morphological Characterization of *Haemonchus contortus* in Sheep (*Ovis aries*) and Goats (*Capra hircus*) from Two Governorates in Yemen

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**Abstract:** This work was conducted to investigate the population and host variation of the parasitic nematode *H. contortus* of sheep and goats from two Governorates in Yemen. Seven morphological characters were investigated namely the number of cuticular ridges (synlophe), morphometrics of total body length, cervical papillae, spicules, barbs, gubernaculum and vulvar flap morphology. Differences were found in total body, cervical papillae and spicule lengths and the numbers of synlophe between the two Governorates. Observations on worm vulval flaps showed that linguiform and knobbed morphs were equally distributed in animals in one Governorate, while the knobbed type was more predominant in the other. It is believed that these variations in worm characteristics between two Governorates may be attributed to geographical isolation and manifestations of some genetic factors during worm establishment and development in the host.

**Key words:** Morphological variations • Morphometrics • Vulval flap morphology

### INTRODUCTION

The large stomach worm, *Haemonchus contortus*, commonly known as "the barber's pole worm", is a blood-sucking nematode found in the abomasums of sheep and goats. Infections can cause anaemia and occasional death of the animals. It is a major animal health problem and can result in severe economic losses due to host morbidity and mortality. In Yemen, both goats and sheep are important meat and milk sources and this worm can cause huge economic losses whenever there is a severe outbreak of infection. In this country, more than 20,000 sheep and 10,000 goats were infected with internal parasites, most of them reported from the two most populated Governorates of Sana'a and Al-Hudaydah [1]. Although the worm from both hosts share similar general morphology, differences in the characteristics of cuticular ridges (synlophe) of *H. contortus* recovered from the two animals have kindled much interest to look further into a more detailed morphology of the worm, more so when *H. contortus* is sympatric in many regions of the world, especially where goats and sheep share the same pastures [2-4].

In the present study, morphometrics of cuticular ridges (synlophe), total body length, cervical papillae of male and female worms, spicules, barbs, gubernaculum of male and female's vulvar flap morphology of *H. contortus* in goats and sheep from two Governorates in Yemen are compared and described.

### MATERIALS AND METHODS

**Study Area:** This study was conducted in two Governorates in Yemen, Sana'a and Al-Hudaydah, which are located in the northern and the western part of Yemen respectively (Fig. 1). These two Governorates were selected over the others as they supported the highest number of sheep and goat populations in Yemen and they are geographically isolated by a chain of mountains.

**Worm Recovery and Parasitological Examination:** Adult *H. contortus* worms were recovered from the abomasa of sheep and goats obtained from the city abattoirs in Sana'a and Al-Hudaydah. The abomasa of 68 sheep and 33 goats from Sana'a and 59 sheep and 105 goats from Al-Hudaydah were collected and examined.



Fig. 1: Map of Yemen Republic showing the location of the study areas (\*).

The abomasa were taken directly to the laboratory, cut open along its greater curvature and individual adult worms were collected and identified under a dissecting microscope. Morphological identification of specimens was referred to Soulsby [5]. The specimens were preserved in 70% alcohol. Observations of body ridges (synlophe) at 8 mm from the anterior end were made on cross-sections of worms obtained by free-hand cuts using sharp razor blades. Specimens were viewed under the interference contrast light microscope using Image Analyzer program. Total body lengths, cervical papillae, spicules, barbs (tip to hook distance), gubernaculum and vulvar flap morphology were observed and measured. Prior to this, each worm specimen was cleared in lactophenol and placed on a glass microscope slide as temporary mounts [4]. One hundred worms from each host for each Governorate, comprising of 50 males and 50 females were randomly chosen for each observation or measurement.

**Statistical Analysis:** Microsoft Excel program was used to store the data and conduct descriptive summary statistics. The software program SPSS 13 (SPSS Inc.) was employed for data analysis and Mann Whitney U-test was used to compare between isolates from the two hosts because the data were non-normally distributed. The significance level for this statistical analysis was set at  $P < 0.05$ .

## RESULTS

Vulvar flap morphology observations showed that linguiform and knobbed morphs were equally distributed in female worms from sheep (42%) in Sana'a whereas the linguiform morph was predominant in goats (50%). In Al-Hudaydah, knobbed females were predominant in the two hosts (Fig. 2).

Morphometric values of *H. contortus* isolates recovered from sheep and goats from the two areas are presented according to sex in Table 1. In Sana'a significant host variation among males of *H. contortus* were found in total body, cervical papillae and right spicule lengths. Males were longer for these variables recovered from goats compared to sheep. In addition the number of cuticular ridges in male worms from goats was higher than males from sheep. Females did not exhibit any significant difference between the two hosts for any of the variable.

However, for Al-Hudaydah, several characteristics were also found to be significantly different between the two hosts. Similar to Sana'a, males recovered from goats were longer in the total body lengths than that recovered from sheep. However, cervical papillae were different between the two hosts in both sexes. In addition, the two spicule characters and synlophe of females showed higher values for goats. Spicules and synlophe measurements of *H. contortus* from Sana'a and Al-Hudaydah are shown in Figs. 3 -10.

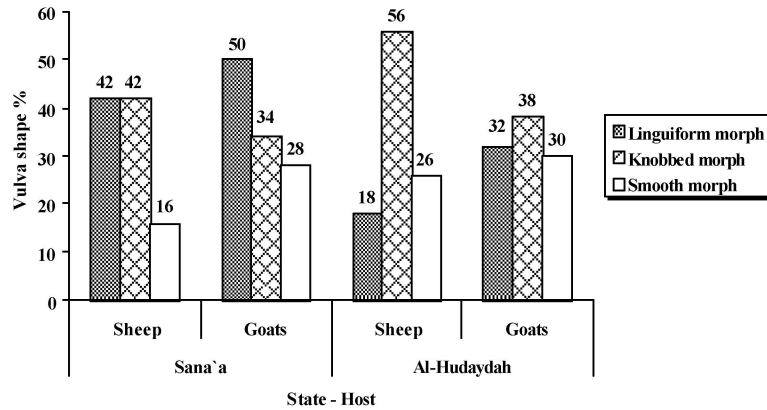


Fig. 2: Distribution of vulvar mrph types of *H. contorts* females in sheep

Table 1. Morphometrics of *Haemonchus contorts* isolates in sheep and goats from Sana'a and Al-Hudaydah\*.

Character	Sex	No.	Sana'a						Al-Hudaydah					
			Sheep		Goats		U-value	P-value	Sheep		Goats		U-value	P-value
			Mean	SD	Mean	SD			Mean	SD	Mean	SD		
BL (mm)	Male	50	17.62	±1.23	18.36	±1.08	809	0.002	16.98	±1.36	18.02	±1.17	741	0.000
	Female	50	28.06	±1.81	27.32	±1.96	987	0.066	26.96	±1.97	27.50	±2.00	1049.5	0.162
CP #	Male	50	412.08	±23.36	422.27	±24.13	965	0.049	393.74	±27.72	417.63	±29.53	739	0.000
	Female	50	437.88	±32.90	426.57	±40.01	1053	0.174	415.57	±35.11	440.73	±31.38	714	0.000
RSL	Male	50	448.40	±22.45	458.32	±19.61	927	0.026	435.71	±24.62	453.01	±20.13	737	0.000
LSL	Male	50	443.83	±23.55	451.09	±20.42	1013	0.102	429.36	±25.43	448.33	±20.43	696	0.000
THr	Male	50	43.79	±3.48	44.36	±4.55	1153	0.504	43.32	±3.67	43.66	±3.32	1171	0.586
THl	Male	50	22.99	±2.32	22.80	±2.60	1188	0.669	22.21	±2.38	22.99	±2.24	1011.5	0.100
GL	Male	50	219.02	±18.35	221.65	±17.36	1123	0.381	211.35	±14.99	215.03	±14.01	1092	0.276
CR †	Male	50	23.38	±1.98	24.22	±1.18	858.5	0.006	22.86	±1.28	22.72	±1.29	1156.5	0.498
	Female	50	23.84	±1.28	24.28	±1.86	1092	0.266	23.74	±1.59	24.88	±1.29	774.5	0.001

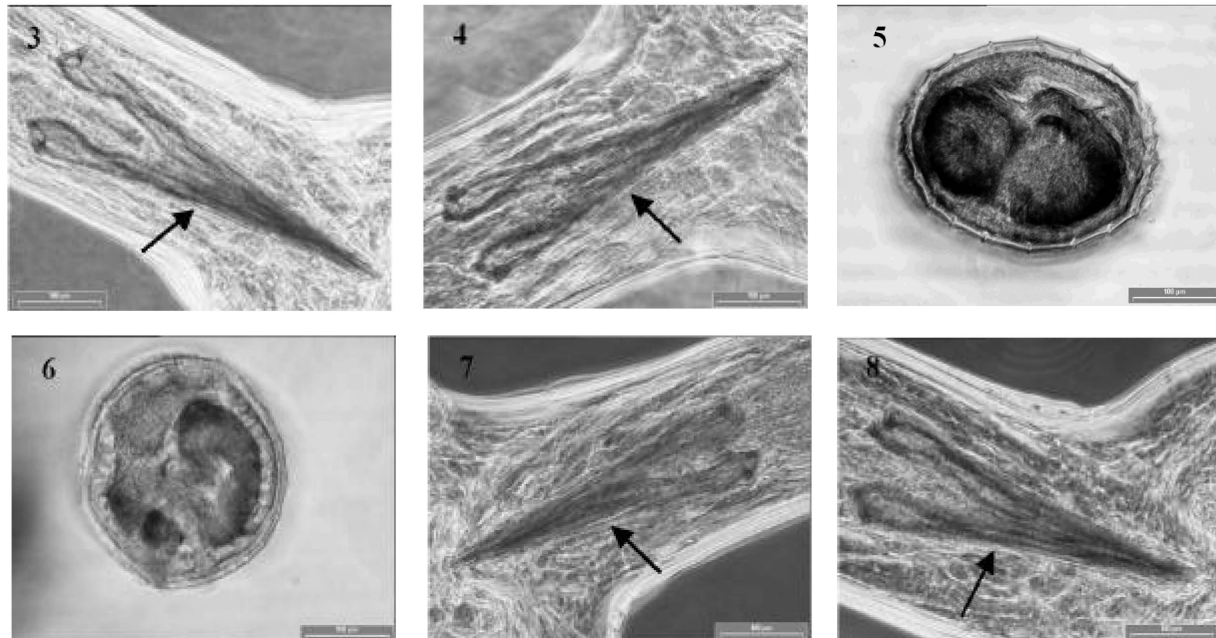
\*: Measurements in micrometers unless noted otherwise

SD: Standard Deviation §: Mann-Whitney-U Test BL: Body length

CP: Cervical papillae #: Measured from anterior extremity RSL: Right spicule length LSL: Left spicule length

THr: Tip-to-hook distance of the right spicule THl: Tip-to-hook distance of the left spicule GL: Gubernaculum

CR: Cuticular ridges †: 8 mm from anterior extremity



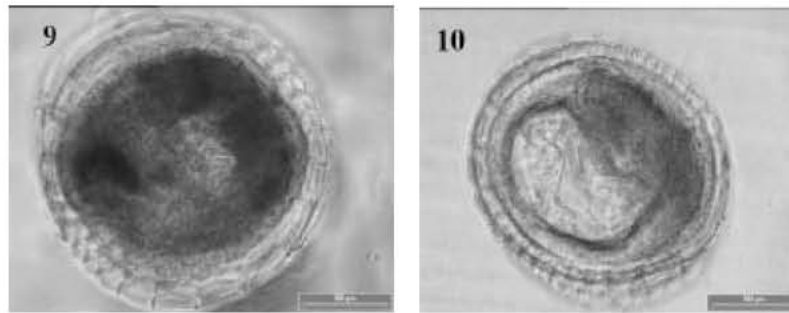


Fig. 3-10: Spicules and synlophe of *H. contortus* recovered from Yemen (scale bar = 100  $\mu$ m).

3. Spicules of *H. contortus* in sheep from Sana'a. 4. Spicules of *H. contortus* in goat from Sana'a. 5 & 6. Cross section at 8mm from anterior end of *H. contortus* female in sheep and goat from Sana'a showing 24, 27 ridges/synlophe respectively. 7. Spicules of *H. contortus* in sheep from Al-Hudaydah. 8. Spicules of *H. contortus* in goat from Al-Hudaydah. 9 & 10. Cross section at 8mm from anterior end of *H. contortus* female in sheep and goat from Al-Hudaydah showing 26, 25 ridges/synlophe respectively.

## DISCUSSION

The present study is the first documentation of morphological variation of *H. contortus* between sheep and goats in Yemen. In this work, morphological differences were found in total body lengths, cervical papillae lengths, spicules lengths and the number of synlophe. Spicule lengths provided the simplest character to use for separating most populations of *H. contortus* and *H. placei* as shown by Lichtenfels *et al.* [3]. In addition, the distance from the tip to the hook of the spicules had also been used as a rapid and easy tool for identification of *Haemonchus* species [6]. The spicules measurements in this study were longer than those reported for *H. contortus* in North America [4].

The distribution of surface cuticular ridges (synlophe) had been used previously to distinguish between populations of *H. contortus* from *H. placei* [2]. Moreover, the number and pattern of longitudinal ridges on the external cuticular surface of trichostrongylid nematodes had been found to be one of the most sensitive and useful character for distinguishing species and determining the relationships among these species [7]. Lichtenfels *et al.* [4] found that the number of cuticular ridges at 10 mm from anterior end of *H. contortus* female was 22 ridges while in the present study, the number of the ridges at 8 mm from anterior end of *H. contorts* female was ranged between 23-24.

The variation in vulvar morphology of female *Haemonchus* worms collected from sheep and goats from the two Governorates may be due to a manifestation of some genetic factors during worm establishment and development in the hosts [8]. However, other authors

considered vulvar morphology as markers of ecological adaptation to an area [9, 10].

In Yemen, sheep and goat populations are higher in Al-Hudaydah and thus would possibly present a wider variation as compared to that in Sana'a'. Also, imports of sheep and goats from Africa and Australia are common, thus may account for the introduction of different strains of *H. contortus* into the country.

On the other hand, variations between isolates of this nematode among locations maybe due to the geographical isolation between the study areas, although how this may affect the infectivity and survival of the worms is not clear. The results of this study also support the possible cross-infection between sheep and goat ruminants, highlighting the importance of defining the role of hosts in the epidemiology of *H. contortus* in Yemen. Thus it is important to have an accurate identification for this worm due to its veterinary and economic importance using the morphological characters which in turn can help in the diagnosis and control of the disease. However, in conclusion, investigation using molecular techniques to study the population genetic structures of *H. contortus* in Yemen has to be carried out in the near future.

## ACKNOWLEDGMENTS

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