



***Haemonchus longistipes* Railliet & Henry, 1909 (Nematoda, Trichostrongyloidea) from dromedary, *Camelus dromedarius* (Artiodactyla, Camelidae), in Egypt: Prevalence and comparative assessment of vulvar morphology**

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Abstract

Examination of 120 abomasa of one-humped dromedary, *Camelus dromedarius*, slaughtered at two slaughterhouses in Egypt during the period from October 2017 to September 2018, revealed that overall 55 (45.83%) dromedaries harbored 12035 *Haemonchus longistipes* worms [5824 (48.39%) males and 6211 (51.61%) females]. Concerning vulvar morphology of female worms, the study revealed that out of 6211 female worms, 515 (43.72%) were smooth, 539 (45.76%) knobbed and 124 (10.53%) linguiform vulvar morphotypes at Elbasateen slaughterhouse (Cairo) and 1555 (30.90%) were smooth, 3049 (60.58%) knobbed and 429 (8.52%) linguiform vulvar morphotypes at Elbakry slaughterhouse (Qalyubia). Accordingly, the knobbed vulvar flap was encountered the most predominant morphotype from dromedaries at both slaughterhouses. Furthermore, the linguiform vulvar flap (L) revealed of 20 (16.13%) LA, 98 (79.03%) LB, 3 (2.42%) LC and 3 (2.42%) LI sub-morphotypes at Elbasateen slaughterhouse and 62 (14.45%) LA, 359 (83.68%) LB, 3 (0.70%) LC and 5 (1.17%) LI sub-morphotypes at Elbakry slaughterhouse. Accordingly, Linguiform B was the most dominant linguiform sub-morphotype from dromedaries at both slaughterhouses. A few numbers of LC and LI might be due to the lower ability of both in coping as compared to LA and LB sub-morphotypes and attributed to adaptation in agro-ecology of the study sites or to the little worms per abomasum in some specimens.

Keywords: one-humped dromedary, *Camelus dromedarius*, slaughterhouse, *Haemonchus longistipes*, vulvar process

Introduction

Gastrointestinal nematode parasitic infection is one of the major health problems in the world ^[1, 2, 3]. Nematode infections affect the health of millions of people and animals, causing a huge economic loss in livestock farming ^[4]. *Haemonchus* spp. (order: Strongylida), which are called the large stomach worms, are pathogenic haematophagous nematodes infecting abomasa of the ruminant hosts, causing significant economic losses worldwide ^[5]. These parasites are gravely problematic in dromedary's husbandry because they not only reduce the productivity and performance of dromedaries, but also predispose them to other infectious diseases ^[6]. Two or sometimes three *Haemonchus* spp. are sympatric in many regions of the world, especially in areas where sheep, goats, cattle and dromedaries are reared together and share the same grazing pasture ^[7, 8]. Cameline haemonchosis, which is caused by *H. longistipes* has an extremely wide geographical range, particularly in tropical and subtropical areas ^[9, 10]. Vulvar process of female *Haemonchus* worms varies in both shape and size ^[11, 12, 13]. Study of the vulvar morphology of female *Haemonchus* worms helps to understand and know more about the worm biology is considered a marker of ecological adaptation ^[12, 14], possesses a great taxonomic importance and determines the type of population that occurs in a certain area ^[13]. Vulvar morphology of *Haemonchus* worms is the manifestation of some genetic factors necessary to establish and develop inside hosts ^[15].

The purpose of this study was to determine: 1- the

prevalence of *H. longistipes* infection among one-humped dromedary, *Camelus dromedarius*, slaughtered at Elbasateen automated slaughterhouse (Cairo) and Elbakry slaughterhouse (Qalyubia). 2- A comparative assessment of vulvar morphotypes of female worms in naturally infected camels.

Materials and methods

Abomasa of 120 one-humped dromedaries were monthly collected from October 2017 to September 2018 from two abattoirs, Elbasateen automated slaughterhouse (Cairo) and Elbakry slaughterhouse (Qalyubia) and transported to the Parasitology Laboratory, Zoology Department, Faculty of Science, Al-Azhar University. Abomasa and its contents were examined, and then, adult *Haemonchus* worms were recovered, according to ^[16]. Worms were extensively washed with physiological saline and stored in 70% ethanol. During the study period, all female *Haemonchus* worms were examined under stereomicroscope to determine the type of cuticular process of vulvar region and classified according to ^[12, 13] to different vulvar morphotypes: linguiform (with a supra vulvar flap), knobbed (with a knob like vulvar process) or smooth (without any vulvar process). Linguiform of female worms were further subclassified into four sub-morphotypes: linguiform A (with one cuticular inflation), linguiform B (without cuticular inflation), linguiform C (with two cuticular inflations) and linguiform I (the cuticular inflation arises from the linguiform process) according to ^[13]. Majority of worms were studied as fresh

samples, whereas some were studied as preserved samples in 70% ethanol and some worms were photographed after clearing in lactophenol and mounting on polyvol [8, 17]. Finally, Microsoft excel was employed for data store and statistical analysis.

Results

Examination of 120 abomasa of one-humped dromedaries, *Camelus dromedarius*, slaughtered in the two slaughterhouses, Elbasateen automated slaughterhouse, Cairo and Elbakry slaughterhouse, Qalyubia revealed that overall 55 (45.83%) dromedary [29 (54.72%) out of 53 from Elbasateen and 26 (38.81%) out of 67 from Elbakry] (Table 1) harbored 12035 *H. longistipes* worms [6211 (51.61%) females and 5824 (48.39%) males] (Table 2). Although dromedaries at Elbasateen slaughterhouse had a high rate of infection with *Haemonchus* worms when

compared with Elbakry slaughterhouse through most months, dromedaries at the second slaughterhouse (Elbakry) had a higher worm abundance than that present infecting dromedaries at Elbasateen slaughterhouse. Vulva was located at the anterior limit of the last third of the female body length. Vulva was either smooth (without a flap), so called smooth vulvar morphotype (Fig. 1), covered with a knob-like flap (knobbed vulvar morphotype) (Fig. 2) or covered with a large and prominent flap (linguiform vulvar morphotype (L)). Further classification of the latter revealed the presence of four sub-morphotypes: LA sub-morphotype (Fig. 3) with vulva covered with a large linguiform flap and had only one cuticular inflation; LB sub-morphotype (Fig. 4) without cuticular inflation; LC sub-morphotype (Fig. 5) with two cuticular inflations in addition to the linguiform flap and LI sub-morphotype (Fig. 6) with one cuticular inflation raised from the linguiform process.

Table 1: Number of infected dromedaries with *H. longistipes*, non-infected and total per month at the two slaughterhouses

Month	Elbasateen slaughterhouse, Cairo			Elbakry slaughterhouse, Qalyubia		
	Positive	Negative	Total	Positive	Negative	Total
October	ND	ND	ND	2	8	10
November	1	1	2	1	7	8
December	3	4	7	1	2	3
January	2	5	7	1	2	3
February	ND	ND	ND	4	6	10
March	4	0	4	4	2	6
April	6	4	10	ND	ND	ND
May	3	2	5	1	4	5
June	5	2	7	2	1	3
July	2	2	4	3	3	6
August	ND	ND	ND	6	4	10
September	3	4	7	1	2	3
Total samples	29	24	53	26	41	67
Total infected %	54.72%			38.81%		

Overall

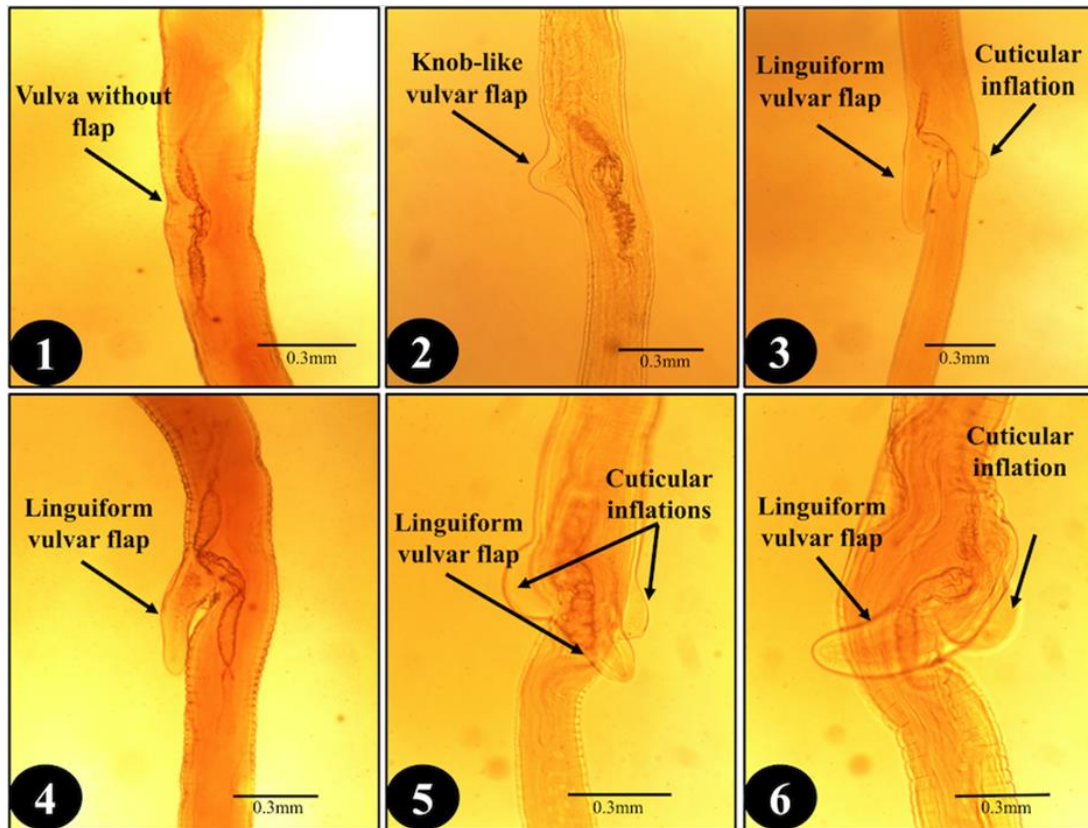
-ND: not done

Table 2: Monthly count and percentage values of male and female worms of *H. longistipes* in dromedaries

Month	Elbasateen slaughterhouse, Cairo				Elbakry slaughterhouse, Qalyubia			
	Male		Female		Male		Female	
	No.	%	No.	%	No.	%	No.	%
October	ND	ND	ND	ND	93	42.27	127	57.73
November	33	54.10	28	45.90	178	43.95	227	56.05
December	42	29.17	102	70.83	3	100.00	0	0.00
January	17	62.96	10	37.04	1	100.00	0	0.00
February	ND	ND	ND	ND	657	45.19	797	54.81
March	131	47.12	147	52.88	32	66.67	16	33.33
April	225	47.67	247	52.33	ND	ND	ND	ND
May	243	41.12	348	58.88	12	52.17	11	47.83
June	298	50.34	294	49.66	1470	46.76	1674	53.24
July	7	77.78	2	22.22	115	41.07	165	58.93
August	ND	ND	ND	ND	2256	52.86	2012	47.14
September	4	100.00	0	0.00	7	63.64	4	36.36
Total worms	1000		1178		4824		5033	

-Total worms of Male 5824 (48.39%) and Female 6211 (51.61%)

-No.: Number of worms; ND: not done



Figs 1-6: Photomicrographs showing the different vulvar morphotypes of female *H. longistipes* in dromedary from the two slaughterhouses 1- Smooth vulvar morphotype (without a flap), 2- Knobbed vulvar morphotype (covered with a knob-like flap), 3- Linguiform vulvar sub-morphotype LA (vulva covered with a large linguiform flap and only one cuticular inflation), 4- Linguiform vulvar sub-morphotype LB (vulva covered with a large linguiform flap and without cuticular inflation), 5- Linguiform vulvar sub-morphotype LC (vulva covered with a large linguiform flap and two cuticular inflation) and 6- Linguiform vulvar sub-morphotype LI (vulva covered with a large linguiform flap and with one cuticular inflation raised from the linguiform process).

On one side, examination of the cuticular process of 6211 female worms revealed the presence of 515 (43.72%) smooth, 539 (45.76%) knobbed and 124 (10.53%) linguiform vulvar morphotypes at Elbasateen slaughterhouse and 1555 (30.90%) smooth, 3049 (60.58%) knobbed and 429 (8.52%) linguiform vulvar morphotypes at Elbakry slaughterhouse (Table 3). The knobbed vulvar flap encountered as the most predominant morphotype in dromedaries at both slaughterhouses. However, there were variations in the proportions of smooth and knobbed morphotypes at both slaughterhouses during the different months of the study period specially from January to May, where smooth morphotype was more than knobbed one. On the other side, the linguiform vulvar flap (L) revealed proportions of 20 (16.13%) LA, 98 (79.03%) LB, 3 (2.42%)

LC and 3 (2.42%) LI sub-morphotypes at Elbasateen slaughterhouse and 62 (14.45%) LA, 359 (83.68%) LB, 3 (0.70%) LC and 5 (1.17%) LI sub-morphotypes at Elbakry slaughterhouse. Accordingly, Linguiform B was the most dominant linguiform sub-morphotype in both slaughterhouses through the study period, followed by LA and a few numbers of LC and LI sub-morphotypes of linguiform vulvar morphotypes. Linguiform A was appeared through the period from February to June at both slaughterhouses and a few numbers through July and August at Elbakry slaughterhouse. Regarding LC and LI were appeared only through June at Elbasateen slaughterhouse, also LC was appeared only through February and LI was appeared through February, June and November at Elbakry slaughterhouse (Table 4)

Table 3: Monthly percentage values of the major vulvar morphotypes of female *H. longistipes* in dromedary

Month	Elbasateen slaughterhouse, Cairo			Elbakry slaughterhouse, Qalyubia		
	Smooth %	Knobbed %	Linguiform %	Smooth %	Knobbed %	Linguiform %
October	ND	ND	ND	14.96	80.31	4.72
November	28.57	71.43	0.00	16.74	80.18	3.08
December	29.41	67.65	2.94	0.00	0.00	0.00
January	60.00	30.00	10.00	0.00	0.00	0.00
February	ND	ND	ND	48.93	38.02	13.05
March	44.22	38.78	17.01	43.75	37.50	18.75
April	42.51	49.39	8.10	ND	ND	ND
May	60.06	27.87	12.07	27.27	18.18	54.55
June	30.95	58.16	10.88	29.27	60.81	9.92
July	50.00	0.00	50.00	40.61	47.27	12.12
August	ND	ND	ND	26.89	67.30	5.82

September	0.00	0.00	0.00	0.00	100.00	0.00
Total worms %	43.72	45.76	10.53	30.90	60.58	8.52
-Total No. of worms	515	539	124	1555	3049	429

ND: not done

Table 4: Monthly percentage values of sub-morphotypes of the linguiform vulvar flap of female *H. longistipes* in dromedary

Month	Elbasateen slaughterhouse, Cairo				Elbakry slaughterhouse, Qalyubia			
	LA%	LB%	LC%	LI%	LA%	LB%	LC%	LI%
October	ND	ND	ND	ND	0.00	100.00	0.00	0.00
November	0.00	0.00	0.00	0.00	0.00	85.71	0.00	14.29
December	0.00	100.00	0.00	0.00	0.00	0.00	0.00	0.00
January	0.00	100.00	0.00	0.00	0.00	0.00	0.00	0.00
February	ND	ND	ND	ND	28.85	68.27	1.92	0.96
March	28.00	72.00	0.00	0.00	0.00	100.00	0.00	0.00
April	15.00	85.00	0.00	0.00	ND	ND	ND	ND
May	4.76	95.24	0.00	0.00	16.67	83.33	0.00	0.00
June	25.00	56.25	9.38	9.38	16.27	81.33	0.60	1.81
July	0.00	100.00	0.00	0.00	10.00	90.00	0.00	0.00
August	ND	ND	ND	ND	1.71	98.29	0.00	0.00
September	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Total worms %	16.13	79.03	2.42	2.42	14.45	83.68	0.70	1.17
- Total No. of worms	20	98	3	3	62	359	3	5

- No.: Number; ND: not done

Discussion

In Egypt, only one study [18] had been carried out on the prevalence of *H. longistipes* worms infecting abomasa of dromedary, *Camelus dromedarius*. The current study revealed that 55 (45.83%) out of 120 abomasa of dromedary in Egypt were infected with *H. longistipes*. This finding was lower than that was previously recorded for dromedaries in Egypt (62%) [18], in Niger (83%) [19] and in Sudan (76.56%) [20]. However, the present finding was greater than the results of [21, 22] in Iran (18% and 14%, respectively). In Pakistan, the infection rate of dromedaries with *H. longistipes* was (52.57%) [23], this result was lower than the infection rate of dromedaries at Elbasateen slaughterhouse (54.72%) but greater than the infection rate of dromedaries at Elbakry slaughterhouse (38.81%). This variation in the results of infection rate might be due to the environmental climate of each locality or country.

Regarding the vulvar flap polymorphism of female *H. longistipes* from dromedaries, no data were recorded except that obtained from [18] in Egypt and [14] in Mauritania, whereas extensively data were available for *H. contortus* infecting small ruminants [14, 24, 25, 26, 27, 28, 29, 30, 31, 32]. The current results revealed the presence of 515 (43.72%) smooth, 539 (45.76%) knobbed and 124 (10.53%) linguiform vulvar morphotypes of *H. longistipes* at Elbasateen slaughterhouse (Cairo) and 1555 (30.90%) smooth, 3049 (60.58%) knobbed and 429 (8.52%) linguiform vulvar morphotypes at Elbakry slaughterhouse (Qalyubia). The knobbed vulvar flap was the most predominant morphotype at both slaughterhouses. These results were in accordance with that recorded for the same *Haemonchus* species from dromedaries in Mauritania [14], but it was in contrast to the finding obtained by [18], who found that linguiform vulvar flap was the predominant morphotype from dromedaries in Egypt. In other *Haemonchus* spp. infecting other hosts, knobbed vulvar flap was the most predominant morphotype of *H. contortus* from sheep in Egypt [30] and goats in Malaysia [31]. Whereas, the predominance of linguiform vulvar morphotype was recorded from sheep in Mauritania [14], in Ethiopia [28, 29, 32] and in Malaysia [31] and from the goats in Ethiopia [32].

In the present investigation, further classification of linguiform vulvar flap revealed the presence of four sub-morphotypes. The study revealed proportions of 20 (16.13%) LA, 98 (79.03%) LB, 3 (2.42 %) LC and 3 (2.42%) LI sub-morphotypes at Elbasateen slaughterhouse (Cairo) and 62 (14.45%) LA, 359 (83.68%) LB, 3 (0.70%) LC and 5 (1.17%) LI sub-morphotypes at Elbakry slaughterhouse (Qalyubia). Accordingly, Linguiform B was the most dominant linguiform sub-morphotype at both slaughterhouses, followed by LA, LI and LC sub-morphotypes except that LC and LI had the same proportion at Elbasateen slaughterhouse. A few numbers of LC and LI might be due to the lower ability of both in coping as compared to LA and LB sub-morphotypes and attributed to adaptation in agro-ecology of the study sites or to the little worms per abomasum in some specimens of the current work. However, this result was in contrast to [18] who found LA was the predominant linguiform vulvar morphotype on the same *Haemonchus* infecting the same host (one-humped dromedary) in Egypt. The current result on *H. longistipes* was in contrast to that recorded for other *Haemonchus* infecting sheep and goats, where, LA was the predominant linguiform vulvar morphotype from sheep in Egypt [30] and from sheep and goats in eastern Ethiopia [32] but in Awassa town, Ethiopia [28] LC was the predominant linguiform vulvar morphotype from sheep and goats.

Variation in vulvar morphology of female *Haemonchus* spp. among different studies might be due to manifestation of some genetic factors during worm establishment and development in the host [15]. Vulvar morphology helps to understand and know more about the biology of *Haemonchus* species and determine the type of population that occurs in sheep, goats and dromedaries in a certain area [13]. Moreover, [14] indicated that vulvar morphology varies with ecology and taken as an adaptation to specific ecology.

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