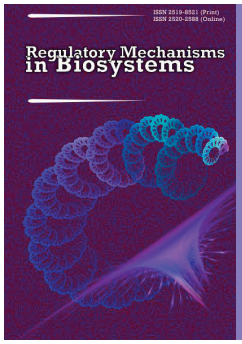

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Differential characters of fleas of the genus *Ctenocephalides* (Siphonaptera, Pulicidae) obtained from dogs

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Fleas are the common name of the order Siphonaptera, obligate provisional nidiculous parasites of mammals and, to a lesser extent, birds, which can also feed on humans. Fleas can cause ctenocephalidosis of dogs and are carriers of many dangerous infectious and invasive diseases. Their ability to use humans as an alternative host determines the importance of these parasites in health care. Therefore, the aim of the work was to establish morphological and metric characters of adult male and female fleas of the species *Ctenocephalides felis* Bouche, 1835 and *C. canis* Curtis, 1826, isolated from domestic dogs for species identification. Morphological studies of fleas have shown that the differential features of *C. felis* and *C. canis* include the shape of the head and anterior-dorsal cuticular notch on the head. Notably, male fleas of both species have a more rounded forehead than female fleas. The metric parameters of adult fleas can be used as additional identification features, which will increase the efficiency of differential diagnosis of parasitic insects. We found differences in male fleas by 23 parameters, of which the value of 17 parameters were smaller in *C. canis* specimens. Males of *C. felis* were 9.1–21.1% larger in total body length, lengths of head and thorax, as well as lengths of mesothorax and metathorax. Differential features included size of the teeth of the main ctenidium and pronotal ctenidium: in *C. felis* males all eight teeth of the main ctenidium, located on the left side, were longer by 12.7–41.7%, and the first, seventh, eighth teeth were wider by 25.4–43.0% than in *C. canis*. In female fleas, differences were found for 24 metric parameters, of which the values of 20 parameters were also smaller in *C. canis* specimens. Females of *C. felis* were 12.1–22.2% larger in terms of total body length, head, breast, prothorax, mesothorax, and metathorax. All their teeth of the main ctenidium were longer by 5.6–40.6%, and the first, second, third, and eighth teeth were 18.1–48.9% wider than in females of *C. canis*. The obtained results add to the already existing data on morphometric features of adult fleas of *C. felis* and *C. canis* species, and will allow timely and accurate diagnosis of ctenocephalidosis in dogs caused by parasites of these species.

Keywords: ctenocephalidosis; ectoparasites; *Ctenocephalides felis*; *Ctenocephalides canis*; identification; morphometry.

Introduction

The entomoses caused by fleas are one of the most common groups of diseases in the population of domestic dogs. The ability to use humans as an alternative host determines the importance of these parasites in health care (De Avelar et al., 2007; Bitam et al., 2010; Dobler & Pfeffer, 2011; Zouari et al., 2017; Ng-Nguyen et al., 2020). Nearly 2000 species of fleas are known by now, grouped into 15 families, which include 200 genera. The most common are fleas of the family Pulicidae, genera *Pulex*, *Ctenocephalides*, *Spilopsyillus*, *Archaeropsyllus* and families Ceratophyllidae, genera *Ceratophyllus*, *Nosopsyllus*, which have important medical significance (Whiting, 2002; Lareschi et al., 2016; Keskin et al., 2018; Galloway, 2019). The genus *Ctenocephalides* Stiles & Collins, 1930 includes 13 species and subspecies, of which only two species of fleas of dogs and cats are prevalent: *Ctenocephalides canis* Curtis, 1826 and *Ctenocephalides felis* Bouche, 1835. *C. felis* has been shown to be the most common species of flea on Earth. It is better adapted to habitat conditions than *C. canis* and, as a result, its history of synanthropic survival has made this geographical species widespread (Beaucournu & Menier, 1998; Rust, 2017). Thus, *C. felis* fleas frequently supplant *C. canis* fleas if both species are found together on dogs. According to scientists, this is due to the fact that *C. canis* is more specific to the host and more demanding of living conditions than *C. felis* (Linardi & Nagem, 1973). However, *C. canis* remains the dominant species in countries such as Korea, Turkey and Greece (Aldemir, 2007; Ahn et al., 2018). *C. felis* is divided into four geographically defined subspecies: cosmopolitan *C. felis felis*, Asian subspecies *C. felis orientis* Jordan, 1925 and two subspecies limited to the African continent, *C. felis* strongylus Jordan, 1925 and *C. felis damarensis* Jordan, 1936. *C. felis* orientis and *C. felis damarensis* have been morphologically recognized as complete species (*C. orientis* and *C. damarensis*, respectively), but the genetic identification of the subspecies *C. felis felis* remains uncertain. Due to the not always characteristic morphological features and the lack of available genetic data for taxa of the genus *Ctenocephalides*, it remains genetically homogeneous in the population of *C. felis* worldwide (Louw al., 1995; Menier & Beaucournu, 1998; Yao et al., 2010; Lawrence et al., 2014; Lawrence et al., 2019).

For fleas of the genus *Ctenocephalides*, the differential morphological features are the presence of well-defined oral ctenidia (combs) and ctenidia located on the back of the pronotum, as well as the presence of bristles on the parietal area of the head. Adult fleas of this genus have a small wingless body, 1–8 mm in size. The proboscis is long, well developed, adapted for skin piercing and blood suction (Ford et al., 2004; Mullen & Durden, 2018). The identification features of the species *C. felis* and *C. canis* include the following features: body colour, head shape, length of teeth of oral ctenidia, the number of bristles on the episternum of the metathorax (metepisternum) and the number of dorsal bristles on hind tibia (Johnson, 1957; Amin & Sewell, 1977). Thus, according to scientific studies, *C. felis* is characterized by a long, sloping forehead. Dorsal thickening of the frontal region of the head is long and narrow. The posterior edge of the hind tibia of this species has one notch where long subapical ventral setae are located. The species *C. felis orientis* is morphologically characterized by a short rounded forehead and a shorter dorsal thickening

compared to *C. felis*. The species *C. canis* is characterized by short, sharply vertical, steep forehead and short dorsal thickening. The ventral edge of the hind tibia has two notches that carry strong setae between the postmedian and subapical setae. *C. felis orientis* can also be distinguished by the presence of a number of tiny setae pointed dorsally from the antennal fossa in females (Hii et al., 2015). Other scientists note that male fleas can be further determined by the shape of manubrium and the size of the edeagus and its hooked end (Holland, 1949; Menier & Beaucournu, 1998). However, despite these differences, some authors suggest additional consideration of chaetotaxia in terms of the number of setae and ctenidia (Amin et al., 1974; Amin, 1976).

Scientists have also noted frequent misidentifications of *C. felis strongylus* and in cases of differentiating *C. canis* from *C. orientis* on the basis of a single criterion, the shape of the head (Beaucournu & Kock, 1990).

Variations in forehead curvature differ significantly in species of the genus *Ctenocephalides*. In *C. felis felis* the forehead is very elongated, and in *C. canis* it is pronouncedly rounded. The number of setae in the occipital region should also be considered: 2 in *C. felis* and *C. orientis*, 3 in *C. canis*, and 1 with micro-setae behind the antenna fossa of females in *C. connatus*. The number of setae on the dorso-ventral side of tibia and lateral metanotal region (LMA) in female fleas of different species varies morphologically: 6 and 2 in *C. felis*, 7 and 2 in *C. orientis*, 8 and 3 in *C. canis*, respectively. Species-specific features in females are also the presence and number of plantar bristles of the fifth tarsomere (2 in *C. felis*, 6 in *C. damarensis* and *C. connatus*) and in the area of the third – sixth sternites (2 in *C. felis*, 3 in *C. connatus*). In male fleas, the authors propose to take into account the shape of manubrium for identification. The manubrium of *C. felis* and *C. damarensis* has a narrowed tip, while that of *C. canis* and *C. orientis* has a widened tip (Lawrence et al., 2019).

In *Ctenocephalides* spp., morphological variations in the structure of ctenidia and chaetotaxia of LMA and hind tibias are often observed. Thus, in Brazil, altered chaetotaxy of LMA or hind tibiae of *Ctenocephalides* spp. were recorded in fleas collected on dogs in the municipality of San Juan del Rei, Minas Gerais and in fleas collected on dogs and cats in the municipality of Rio de Janeiro (Fernandes et al., 1996). Other researchers have also found variations in chaetotaxy of *Ctenocephalides* spp. Namely, the fluctuations in the number of setae of LMA or metatibia were found in 54.5% of *C. felis* (Rodrigues et al., 2001; Stalliviere et al., 2009). Of the 87 studied specimens of *C. felis*, 31.0% showed variations in the number of bristles of LMA and hind tibiae. Moreover, variations were found in 40.7% of female specimens and in 15.1% of males, and the difference between indicator values was significant (Linardi & Santos, 2012).

Thus, the morphological identification of fleas has become increasingly significant due to their important role in the accumulation and transmission of pathogens of dangerous bacterial, viral and invasive diseases. Identification of parasitic insect species is necessary in order to make objective scientific conclusions about their physiological, ecological and zoogeographical properties, which have epizootiological and epidemiological significance. The identification of flea species is one of the primary and necessary stages of a comprehensive study in the effective control and prevention of parasitism of *Ctenocephalides* spp.

The aim of the work was to find morphological and metrical parameters of adult stages of development of male and female fleas for identification of *C. canis* and *C. felis*, isolated from domestic dogs.

Materials and methods

The study was performed during 2016–2020 on the basis of the laboratory of the Department of Parasitology and Veterinary Sanitary Examination of the Faculty of Veterinary Medicine of Poltava State Agrarian Academy and at the veterinary practice "Vetexpert" (Poltava).

Fleas were collected from the dogs admitted to the veterinary practice "Vetexpert" by combing with a plastic small-toothed special comb for 10 minutes. The insects were fixed in 70% ethyl alcohol. Species of isolated parasitic insects were identified by morphological taxonomic features according to keys (Ioff & Skalon, 1954; Ioff et al., 1965; Wall & Shearer, 2001) using microscopy. 260 specimens of *C. felis* (60 males and 200 females), and 137 specimens of *C. canis* (30 males and 107 females) were subjected to morphological analysis.

In order to study the morphometric differential parameters of fleas of the identified species, permanent slides were prepared according to the generally accepted method, fleas were placed on the left side (Tiflov et al., 1977). Metric parameters were measured using ImageJ for Windows® software (version 2.00) in interactive mode using a lens ×5, ×10, ×40 and a photo eyepiece ×10. Microphotography was performed using a digital camera with a microscope Sigeta M3CMOS 14000 14.0 MP (China).

Standard deviation (SD) and average values (x) were calculated. Significance of difference between average values in the studied adult male and female fleas of the species *C. canis* and *C. felis* was established using one-way analysis of variance and F-test for P = 0.05 confidence level.

Results

C. felis and *C. canis* fleas have a typical morphological structure for insects, with some differences specific of the genus *Ctenocephalides*. Those fleas are wingless parasitic insects. The body of the imago is covered by cuticle strongly flattened laterally, and is anatomically divided into three tagmata: head, chest and abdomen (Fig. 1).

Each tagma is characterized by certain structural features and has different formations of various functional purpose. Also characteristic of fleas of the genus *Ctenocephalides* is the presence of ctenidia, the series of teeth that form well-defined ridges. They are located in the lower part of the insect's head (oral ctenidia) and on the back of prothorax (pronotal ctenidia). The flea thorax consists of three segments, namely protorax, mesothorax and metatorax. In each of these segments there are dorsal half-rings: pronotum, mesonotum and metanotum. On the sides of each thoracic segment there are the lateral plates (propleura, mesopleura and metapleura). Three pairs of legs are attached to each thoracic segment. Each leg consists of a coxa, trochanter, femur, tibia, and tarsus which has 5 joints and ends with a pair of claws. The flea abdomen contains 10 segments, each of which consists of dorsal (tergite) and abdominal (sternite) semirings.

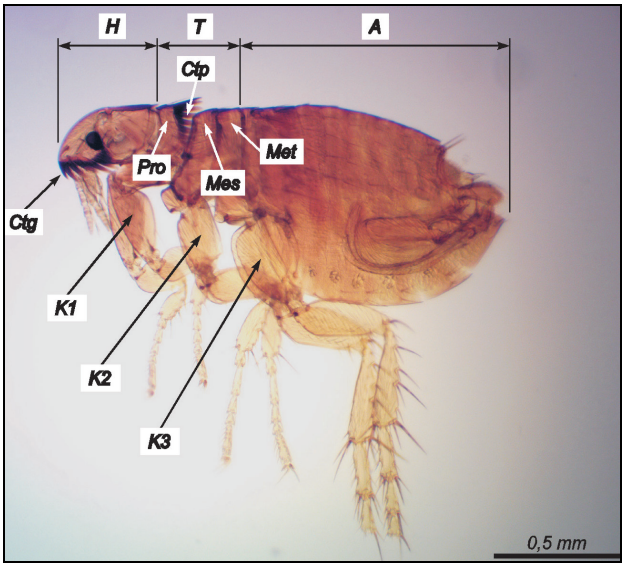

Fig. 1. General morphology of adult *Ctenocephalides* spp. fleas: *H* – head, *T* – thorax, *A* – abdomen, *Ctp* – pronotal ctenidium, *Ctg* – genal ctenidium, *Pro* – pronotum, *Mes* – mesonotum, *Met* – metanotum, *K1*, *K2*, *K3* – coxa I, II and II pairs of legs

The head of fleas of the genus *Ctenocephalides* is divided by the antennal fossa into the frontal and posterior (occipital) parts (Fig. 2). In the frontal part of the head there are the forehead, mouth, genae, gulae, eyes and bristles. The eyes are large round-oval, dark, well visible. There is an antenna in the antennal fossa, it consists of three segments: the first, which is the main segment, the second segment bears long bristles, the third is a club. The last segment is subdivided into a number of sub-segments. Moreover, the front part of the antennal fossa is covered by the edge of gula with a false tooth at the end. The mouth apparatus of piercing-sucking type in parasitic insects is located in the lower part of the front of the head. The external oral organs of fleas consist of several parts (Fig. 3).

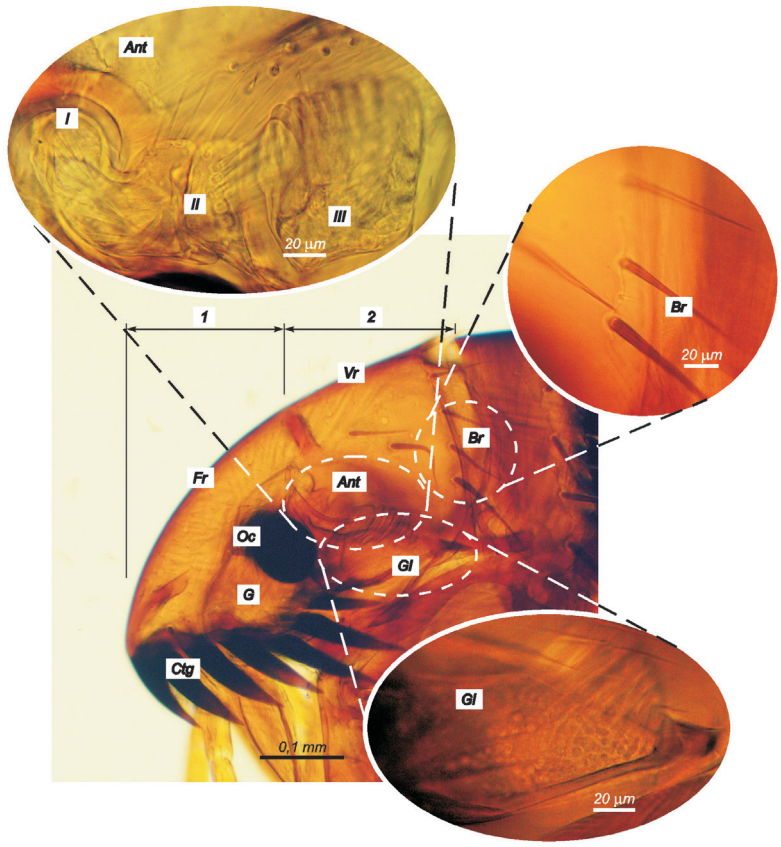

Fig. 2. Morphology of the head tagma of adult *Ctenocephalides* spp.: *1* – frontal part of head, *2* – occipital part of head; *Vr* – vertex, *Fr* – frons, *G* – gena, *Ant* – antenna, *Gl* – gula, *Oc* – ocella, *Ctg* – genal ctenidium, *Br* – row of bristles at vertex; *I* – first segment of antenna, *II* – second segment of antenna, *III* – third segment of antenna (club)

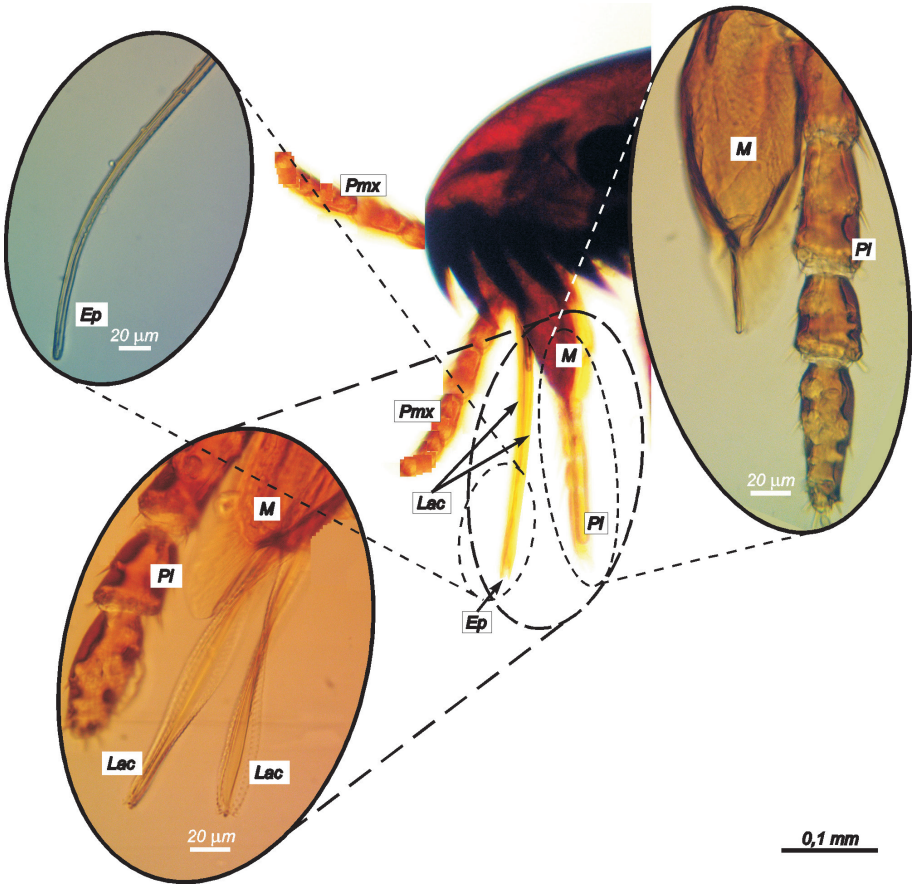

Fig. 3. Morphology of mouth apparatus of adult *Ctenocephalides* spp.: *Ep* – epipharynx, *Lac* – lacinia (maxillary stylets), *Pmx* – maxillary palps, *Pl* – labial palps, *M* – maxillae

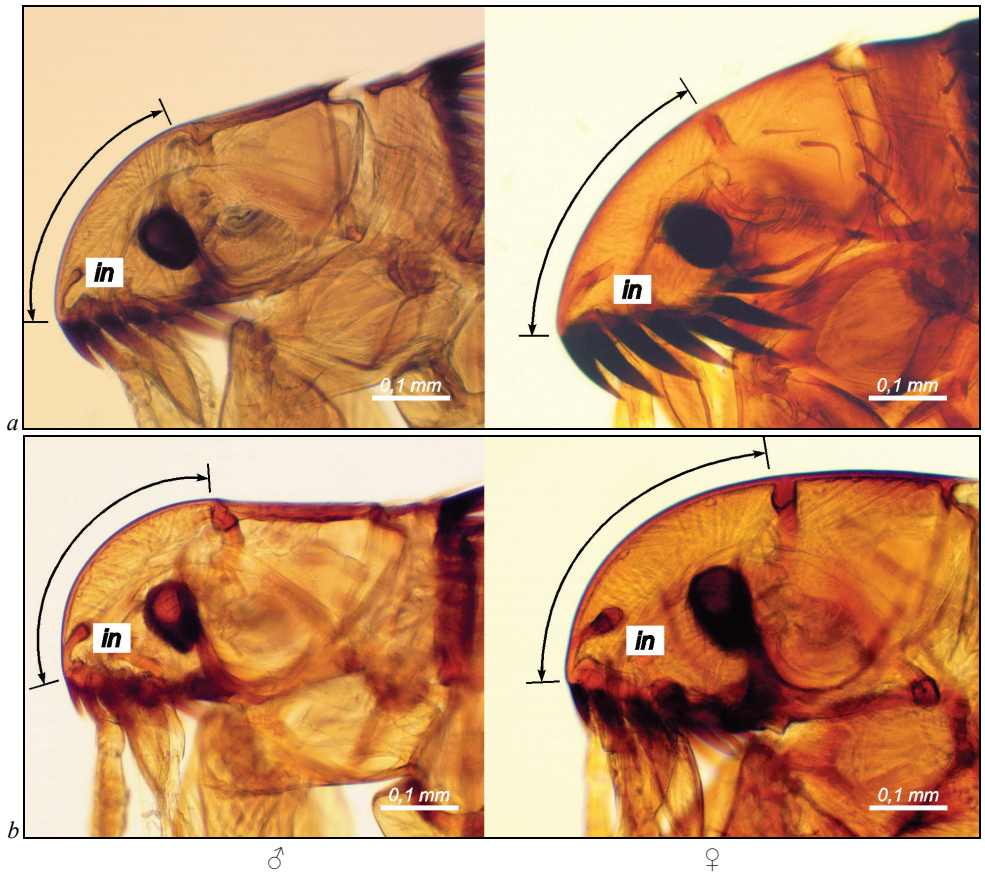

Fig. 4. Specifics of frontal part of the head of adult fleas (lateral view): *a* – *Ctenocephalides felis*, *b* – *C. canis*; *in* – dorsal incrassation

The unpaired piercing organ is represented by the epipharynx (labrum), shaped as a narrow long stiletto. The paired prickly organ is represented by the upper jaws (mandibles, lacinia), which are covered with small teeth and also have the shape of sharp stilettos. Together, all three organs (epipharynx, lacinia) form a tube for blood suction. Behind the piercing parts of the flea's oral apparatus are the labium and maxillae. Moreover, the labium has two articulated processes, the maxillary palps, which when folded, serve as a case for the piercing organs and all these formations form a rostrum. Two flat maxillae have a triangular shape and carry articulated maxillary palps, which are most visible of all oral organs.

It can be noted that the differential species differences of *C. felis* and *C. canis* include the shape of the head and dorsal incrassation on its anterior part (Fig. 4). Thus, in *C. felis* fleas, the forehead is more elongated, sloping, and the incrassation is longer and thinner (Fig. 4a). At the same time, in *C. canis* fleas, the forehead is short and abruptly rounded, and the incrassation is thick and club-shaped (Fig. 4b). Moreover, male adults of fleas of both species have a more rounded forehead than females.

The number of teeth on the head and pronotal ctenidia is the same in fleas of *C. felis* and *C. canis*. At the same time, their number differs between males and females. Thus, in adults of both species, the number of ctenidia teeth located on the head is 16. However, the number of teeth of pronotal ctenidia ranges 14–15 in males, and 15–17 in females. Significant differences were established in the metric parameters of adult *C. felis* and *C. canis* fleas for both males and females. In males of both species morphometric differences were determined by 23 parameters, and the values of 17 of those parameters were lower in *C. canis* (Table 1).

The body of *C. felis* male fleas was longer by 9.2 % (2.38 ± 0.12 mm, $P < 0.05$) than that of *C. canis*. The head and thorax were also longer in *C. felis* fleas by 9.1% (0.44 ± 0.02 mm, $P < 0.01$) and 16.1% (0.47 ± 0.08 mm, $P < 0.01$), respectively, than the same parameters in *C. canis* fleas. The meso- and metathorax were longer in *C. felis* by 21.1% (0.19 ± 0.01 mm, $P < 0.001$) and 18.2% (0.22 ± 0.01 mm, $P < 0.001$), respectively, compared with *C. canis*. The measurements of the genal ctenidium can be considered among differential species characters, because they were significantly different in *C. felis* and *C. canis*.

Table 1
Metric parameters of ♂ fleas of the genus *Ctenocephalides*, obtained from dogs (n = 10)

Parameters		*C. felis*		*C. canis*	
		x ± SD	min–max	x ± SD	min–max
Length of body, mm		2.38 ± 0.12	2.23–2.64	2.16 ± 0.25*	1.73–2.50
Leangth of head, mm		0.44 ± 0.02	0.41–0.46	0.40 ± 0.02**	0.36–0.44
Length of thorax, mm:		0.56 ± 0.03	0.52–0.61	0.47 ± 0.08**	0.36–0.58
– prothorax		0.15 ± 0.01	0.12–0.16	0.13 ± 0.03	0.09–0.17
– mesothorax		0.19 ± 0.01	0.17–0.21	0.15 ± 0.03***	0.12–0.19
– metathorax		0.22 ± 0.01	0.20–0.24	0.18 ± 0.03***	0.15–0.22
Length of abdomen, mm		1.39 ± 0.10	1.28–1.58	1.30 ± 0.19	0.98–1.55
Teeth of genal ctenidium, μm:					
I	– length	82.8 ± 2.1	80.1–86.0	58.2 ± 4.9***	50.4–62.6
	– width	15.4 ± 0.9	14.0–16.6	11.5 ± 1.2***	9.1–12.9
II	– length	120.1 ± 1.7	117.9–122.7	84.3 ± 1.7***	82.1–87.8
	– width	18.5 ± 1.0	17.3–20.1	20.9 ± 1.6*	18.9–23.2
III	– length	127.4 ± 1.2	125.3–128.9	106.5 ± 4.1***	101.0–117.0
	– width	23.6 ± 0.7	22.3–24.4	26.5 ± 1.7***	23.1–28.4
IV	– length	127.0 ± 1.1	125.3–128.5	110.6 ± 2.3***	105.0–112.6
	– width	24.2 ± 0.8	23.2–25.6	36.5 ± 4.4***	28.3–41.9
V	– length	134.2 ± 3.1	130.0–137.4	115.9 ± 1.9***	112.8–118.4
	– width	24.1 ± 0.9	22.9–25.2	28.8 ± 1.1***	27.1–30.4
VI	– length	130.4 ± 2.0	128.3–134.9	92.8 ± 1.5***	90.2–95.4
	– width	21.8 ± 1.0	20.2–23.1	23.9 ± 1.8**	20.5–26.2
VII	– length	89.0 ± 2.5	82.7–91.3	51.9 ± 1.7***	49.6–54.9
	– width	16.7 ± 0.7	15.3–17.4	12.0 ± 0.7***	11.0–13.1
VIII	– length	50.8 ± 2.5	47.4–54.3	31.6 ± 1.2***	30.1–33.6
	– width	8.4 ± 0.6	7.2–9.2	4.8 ± 0.4***	4.1–5.1
Length of teeth of pronotal ctenidium, μm:					
– dorsal		142.8 ± 4.1	138.0–148.7	134.5 ± 2.8***	131.0–140.3
– terminal		59.1 ± 1.1	57.4–60.3	65.2 ± 5.1***	59.6–72.4

Note: * – $P < 0.05$; ** – $P < 0.01$; *** $P < 0.001$ – compared with values of parameters of *C. felis*.

Thus, in *C. felis* fleas all 8 teeth were longer by 12.7–41.7% (from 50.8 ± 2.5 to 134.2 ± 3.1 μm, $P < 0.001$) compared to the same teeth in *C. canis*. Also, the number of genal ctenidium teeth were wider in *C. felis*

fleas: first tooth was by wider by 25.4% (15.4 ± 0.9 μm, P < 0.001), seventh tooth by 27.8% (16.7 ± 0.7 μm, P < 0.001), and eighth tooth by 43.0% (8.4 ± 0.6 μm, P < 0.001) compared to those of *C. canis*. The second, third, forth, fith and sixth teeth were narrower in *C. felis* fleas by 8.8–33.6% (from 18.5 ± 1.0 to 24.2 ± 0.8 μm, P < 0.001–0.05) compared with *C. canis*. The dorsal tooth of the pronotal ctenidium was longer by 5.8% (142.8 ± 4.1 μm, P < 0.001), and the terminal tooth of the pronotal ctenidium was shorter by 9.4% (59.1 ± 1.1 μm, P < 0.001) in *C. felis* fleas compared with *C. canis*.

Twenty five metric parameters of *C. felis* and *C. canis* female fleas were analyzed; by twenty four of those, the species significantly differed (Table 2). *C. felis* female fleas were larger than *C. canis* fleas by 20 morphometric parameters. In contrast, *C. canis* female fleas were larger than those of *C. felis* by 4 morphometric parameters.

Table 2

Metric parameters of ♀ fleas of the genus *Ctenocephalides*, obtained from dogs (n = 10)

Characters	*C. felis*		*C. canis*	
	x ± SD	min–max	x ± SD	min–max
Length of body, mm	3.58 ± 0.17	3.28–3.76	3.06 ± 0.17***	2.83–3.37
Length of head, mm	0.56 ± 0.04	0.51–0.61	0.46 ± 0.03***	0.40–0.49
Length of thorax, mm:	0.71 ± 0.08	0.53–0.80	0.57 ± 0.05***	0.52–0.68
– prothorax	0.21 ± 0.03	0.16–0.24	0.17 ± 0.02**	0.15–0.21
– mesothorax	0.24 ± 0.03	0.18–0.27	0.19 ± 0.02***	0.16–0.23
– metathorax	0.27 ± 0.03	0.19–0.29	0.21 ± 0.02***	0.19–0.24
Length of abdomen, mm	2.31 ± 0.14	2.16–2.52	2.03 ± 0.16***	1.79–2.24
Teeth of genal ctenidium, μm:				
I – length	120.3 ± 6.7	109.5–128.9	71.4 ± 1.3***	70.1–74.8
I – width	21.1 ± 1.4	18.2–23.2	13.0 ± 1.0***	11.2–14.2
II – length	149.6 ± 2.3	145.0–153.9	91.4 ± 1.4***	89.5–93.7
II – width	28.9 ± 1.2	26.4–30.3	23.7 ± 1.6***	21.3–25.9
III – length	153.3 ± 2.0	150.2–155.2	118.2 ± 4.2***	109.0–122.0
III – width	36.0 ± 1.3	33.5–37.8	27.3 ± 1.2***	25.0–29.1
IV – length	146.7 ± 0.9	145.2–148.0	121.2 ± 2.3***	118.3–124.7
IV – width	31.4 ± 1.4	30.1–34.2	37.6 ± 1.2***	36.1–39.5
V – length	149.2 ± 1.4	147.1–151.0	123.1 ± 2.0***	120.5–125.9
V – width	29.8 ± 1.6	27.1–32.0	32.2 ± 1.9**	30.2–35.3
VI – length	147.4 ± 1.9	144.9–150.0	123.0 ± 3.2***	119.9–127.9
VI – width	26.3 ± 1.3	24.7–28.2	29.2 ± 1.6***	26.1–31.0
VII – length	97.1 ± 4.4	88.5–101.3	91.7 ± 2.7**	87.7–96.1
VII – width	18.8 ± 0.9	17.0–20.1	17.8 ± 1.8	15.3–20.2
VIII – length	63.3 ± 5.1	50.5–68.2	54.8 ± 3.8***	48.7–60.2
VIII – width	16.7 ± 2.1	13.1–19.4	8.6 ± 1.6***	6.9–11.1
Length of teeth of the pronotal ctenidium, μm:				
– dorsal	156.4 ± 4.2	148.3–162.2	151.4 ± 1.9**	148.2–154.3
– terminal	83.8 ± 3.5	79.0–89.2	89.7 ± 1.9***	85.5–92.2

Note: ** – P < 0.01; *** – P < 0.001 – compared to the values of parameters of *C. felis*.

The body and tagmata (head, thorax, abdomen) of *C. felis* females were longer by 12.1–19.7% (from 0.56 ± 0.04 to 3.58 ± 0.17 mm, P < 0.001) than in *C. canis*. The length of the pro-, meso- and metathorax of *C. felis* fleas was also greater by 19.1% (0.21 ± 0.03 mm, P < 0.001), 20.8% (0.24 ± 0.03 mm, P < 0.001) and 22.2% (0.27 ± 0.03 mm, P < 0.001) than in *C. canis*. In *C. felis* female fleas, all teeth of the genal ctenidium were longer by 5.6–40.6% (from 63.3 ± 5.1 to 153.3 ± 2.0 μm, P < 0.001–0.01) than in *C. canis* fleas.

At the same time, in *C. felis* fleas the following teeth of the genal ctenidium were wider compared with *C. canis*: the first by 38.4% (21.1 ± 1.4 μm, P < 0.001), the second by 18.1% (28.9 ± 1.2 μm, P < 0.001), the third by 24.4% (36.0 ± 1.3 μm, P < 0.001) and the eighth by 48.9% (16.7 ± 2.1 μm, P < 0.001). The fourth, fifth, and sixth teeth, in contrast, were narrower in *C. felis* by 7.4–16.6% (26.3 ± 1.3 to 31.4 ± 1.4 μm, P < 0.001–0.01) compared to *C. canis*. The dorsal tooth of the pronotal ctenidium in *C. felis* fleas was 3.2% longer (156.4 ± 4.2 μm, P < 0.01), and the terminal, on the contrary, was shorter by 6.6% (83.8 ± 3.5 μm, P < 0.001) compared to parameters of *C. canis*.

The obtained data on the metrical parameters of *C. felis* and *C. canis* will help increase the efficiency of the species identification for both males and female fleas.

Discussion

A review of scientific studies shows that ctenocephalidosis of dogs, caused by the parasitism of *C. felis* and *C. canis* fleas, is recorded on most continents. In some regions, the infestation of dogs with blood-sucking parasites ranges from 1.4% to 100%. Moreover, the rates of flea infestation depend on the dogs' age, breed, housing conditions, climatic conditions and preventive and hygienic measures (Linardi & Nagem, 1973; Alcaíno et al., 2002; Linardi & Santos, 2012; Salant et al., 2014). In particular, ctenocephalidosis was found in 49.5% of examined dogs in Poltava city (Ukraine): *C. felis* fleas were dominant with P of 36.1%, *C. canis* were found less often with P of 27.9% (Yevstafieva et al., 2020). This high prevalence causes significant interest in these parasitic insects among scientists. Interest in the study of the morphological identification of fleas has also recently increased due to the fact that they play an important role in the storage and transmission of dangerous pathogens of bacterial, viral diseases and infestations.

Species identification of parasitic insects is necessary in order to make objective scientific conclusions about their physiological, ecological and zoogeographical properties, which has epizootological and epidemiological significance (Zouari et al., 2017; Ng-Nguyen et al., 2020).

We have shown that *C. felis* and *C. canis* fleas have a morphological structure which is common for insects with the morphological differences characteristic of the genus and the species. Species differences include the shape and frontal dorsal incrassation of the head. In particular, in *C. felis* fleas, the forehead is more elongated, sloping, and the incrassation is longer and thinner. In *C. canis* fleas, the forehead is short and steeply rounded, and the incrassation is thick and club-shaped. Notably, the adult males of both flea species have a more rounded forehead than females.

The obtained data on the species morphological identification of obtained fleas are consistent with the results of most studies, which indicate that in *C. felis* the forehead is most elongated, and in *C. canis* it is distinctly rounded (Hii et al., 2015; Lawrence et al., 2019). A number of authors note that the fleas of the genus *Ctenocephalides* can be erroneously identified if only the shape of head is considered (Beaucournu & Kock, 1990). Thus, scientists have proposed to take into account the number of ctenidia and the number of bristles in different parts of the body, especially in LMA (Johnson, 1957; Amin & Sewell, 1977; Lawrence et al., 2019). However, there are reports that indicate that in *Ctenocephalides* spp., morphological variations in the structure of ctenidia and chaetotaxia on LMA are often observed (Fernandes et al., 1996; Rodrigues et al., 2001; Stalliviere et al., 2009; Linardi & Santos, 2012).

Also, our research revealed that the number of teeth on the genal and pronotal ctenidia of *C. felis* and *C. canis* fleas is the same. The number of teeth of the genal ctenidium is 16. The number of teeth of the pronotal pronotum in males ranges from 14 to 15, and in females from 15 to 17. Therefore, it is impractical to take into account the number of teeth of ctenidia in the species identification of *C. felis* and *C. canis*. We analyzed the metric parameters of adult males and females of *C. felis* and *C. canis* in order to increase the efficiency of their species identification. When comparing the metric parameters of adult males of *C. felis* and *C. canis* fleas, significant differences were found in 23 parameters. *C. canis* fleas were smaller by 17 of these parameters by 5.8–43.0% (P < 0.001–0.05), namely: total body length, length of the head and thoracic tagmata, length of meso- and metathorax, length of teeth of the genal ctenidium, width of the first, seventh, eighth teeth of the genal ctenidium, length of the dorsal tooth of the pronotal ctenidium. Male fleas of the same species were larger by 6 parameters by 8.8–33.6% (P < 0.001–0.05) than those of *C. felis*, namely: the width of the second, third, fourth, fifth, sixth teeth of the genal ctenidium, the length of the terminal tooth of the pronotal ctenidium.

When comparing the metric parameters of adult female fleas of the species *C. felis* and *C. canis*, significant differences were found by 24 parameters. *C. canis* fleas were smaller by 3.2–48.9% (P < 0.001–0.01) by 20 of those indicators, namely: total body length, length of the head, thorax and abdomen, length of pro-, meso- and metathorax, length of teeth of the genal ctenidium, width of the first, second, third, eighth teeth of the genal ctenidium, length of the dorsal tooth of the pronotal ctenidium. Female *C. canis* fleas were significantly larger by 6.6–16.6% (P < 0.001–0.01) only by 4 indicators compared to *C. felis*, namely: width of the

fourth, fifth, sixth teeth of the genal ctenidium, and length of the terminal tooth of pronotal ctenidium.

The obtained data on the features of morphometric structure of adult male and female fleas of the species *C. felis* and *C. canis* expand the existing basis of their identification and allow timely and accurate diagnosis of ctenocephalidosis in dogs caused by parasitic insects of these species.

Conclusion

The studies revealed differential morphometric features of the adult male and female fleas of *Ctenocephalides felis* (Bouche, 1835) and *C. canis* (Curtis, 1826), which parasitize domestic dogs. It is proposed to use 23 morphometric parameters to identify male adults, by 17 of which *C. canis* fleas are smaller compared to *C. felis*. The parameters include the total length of the body and its tagmata (head, chest, meso- and metathorax), the length and width of the teeth of the genal and pronotal ctenidia. 24 morphometric parameters are proposed to identify adult females. By 20 of these parameters, *C. canis* fleas are smaller. These indicators include the total length of the body and tagmata (head, thorax, prothorax, mesothorax and metathorax, abdomen), as well as the length and width of the teeth of genal and pronotal ctenidia.

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