

Short communication

Comparative study of the claws of *Pediculus humanus capitis* between archaeological and modern specimens



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ABSTRACT

Metric data of the claws of archaeological specimens of *Pediculus humanus capitis* (dating between 1500 B.C. and A.D. 1500) and modern lice specimens coming from school children were analyzed and compared. Both sets of samples come from Arica in northern Chile. The overall sample is comprised of 14 archaeological specimens (6 females and 8 males) of *Pediculus humanus capitis* and 22 modern specimens (13 females and 9 males). All specimens were studied with scanning electron microscopy (SEM), uncoated, using variable pressure mode. The objective of this study was to metrically analyze the first couple of clutches of ancient and modern adult lice specimens (width and length of the tibio-tarsal claw and tarsus length) to test if morphological changes have taken place throughout time in these anatomical elements.

We found that archaeological male and female specimens presented significant differences in the tibio-tarsal width (right and left). When comparing data between archaeological and modern male specimens, statistically significant differences were found in almost all the parameters studied, except for the right tarsal length. On the other hand, archaeological and modern female specimens showed no statistically significant change in the variables studied. In brief, our data suggest that modern male specimens have undergone a process of claw reduction, but females have maintained the same dimensions.

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1. Introduction

Pediculus humanus capitis, causal agent of pediculosis, is an insect of the suborder Anoplura including the Pediculidae Family with the genera *Pediculus humanus var corporis* and *Pediculus humanus capitis var* (Linnaeus 1758) and Phthiridae family with the genera *Phthirus pubis* (Linnaeus, 1758) (Espinosa and Vázquez, 2002). This species does not recognize geographical, socio-economic, or cultural barriers and it is probably one of the oldest ectoparasites of humanity (Boutellis et al., 2014; Espinosa and Vázquez, 2002; Retana-Salazar, 1994). Its morphology and genomic variation allow for discussion on our evolutionary pass and the peopling of the Americas (Retana-Salazar, 2005; Weiss, 2009).

Pediculus humanus capitis (*P. h. capitis*) feed directly from venules, has a simple metamorphosis, and three life stages: egg, nymph and adult (Espinosa and Vázquez, 2002). *P. h. capitis* has no

wings, but has a long head with a keratinized body. Its flattened back-ventral body allows the ectoparasite to move easily between the hairs of the host and makes it difficult to be caught by combs (Lehane, 2005). Its sclerotized exoskeleton, particularly in the tibia, prevents shrinkage from drying by the passage of time, allowing comparative analysis with modern specimens (Burgess, 1995).

It is well known that *P. h. capitis* females are larger than males. The female's length ranges from 2.4 to 4 mm, while males are between 2.3–3 mm (Zúñiga and Caro, 2010). In this study, we focus instead on the claw structure as it has received less attention. *P. h. capitis* has 3 pairs of legs and each one is composed of six segments: coxa, trochanter, femur, tibia, tarsus, and pretarsus or claw (Keilin and Nuttall, 1930) (Fig. 1). The tarsus has the tarsal nail which can be folded on the tibia forming a curved claw. This structure is associated with sexual dimorphism. The claw is wider in males and has a prominence that is similar to a thumb on the tibial segment, which is used during intercourse, a morphological feature that is absent in females (Villalobos et al., 1998). Ramírez-Morales (2006) indicates that there is a direct relationship between the tibio-tarsal complex and the type of substrate to which lice are held. This selection mainly affects the length of tibial nail and tarsal nail, and also

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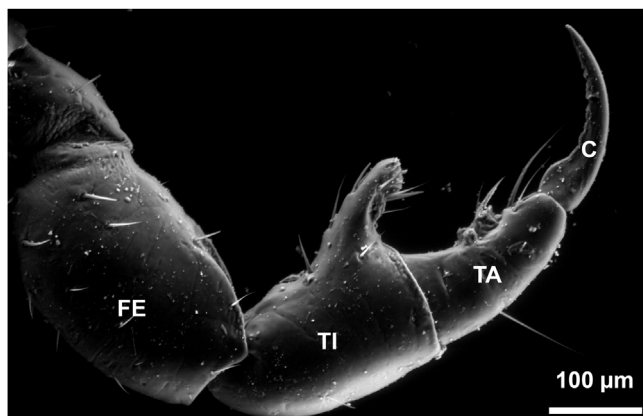


Fig. 1. Anatomical segments of the last portion of the first pair of *Pediculus humanus capitis* leg. FE: Femur, TI: Tibia, TA: Tarsus, C: Claw.

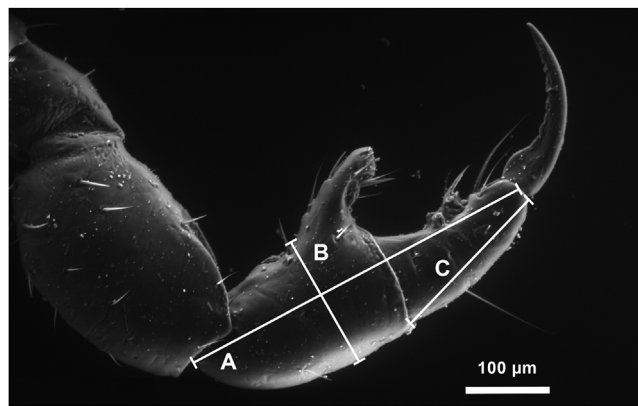


Fig. 2. Measurements taken; (A) Tibio-tarsal length; (B) Tibio-tarsal width; (C) Tarsal length.

has taxonomic importance for segregating different species of lice (Retana-Salazar and Ramírez-Morales, 2006). Thus, the aim of our study is to compare the dimensions (width and length) of the tibio-tarsal claws on the first pair of legs of archaeological specimens with modern specimens of *P. h. capitis* to explore anatomical changes through time. Considering that archaeological lice specimens are about three thousand years old, and that the thickness of hair varies according to the population to which the louse must adapt, and that modern Arica populations have undergone miscegenation, we hypothesize that there should be important morphological changes in the first pair of claws of archaeological specimens compared to modern specimens.

2. Materials and methods

Thirty-six adult specimens of *P. h. capitis*, of which 14 were archaeological (6 females and 8 males) and 22 modern (13 females and 9 males) were analyzed. Archaeological samples were collected

directly from the hair of naturally mummified bodies from four archaeological sites: Quiani-7 (n = 4); Tarapaca-40A (n = 4). Azapa-140 (n = 4) and Camarones-9 (n = 2), all located in the north of Chile (Arica). All the mummies were housed at the Arica, Universidad de Tarapacá Archaeology Museum. The first archaeological site corresponds to people who subsisted on fishing and gathering at the end of Archaic Period (1300–1500 B.C.). The second site dates to the Formative Period (1000 B.C.–A.D. 500) and the third and fourth sites date to the Late Intermediate and Late Periods, and are associated with mixed economic subsistence practices (fishing and farming) (A.D. 1400–1500).

Modern lice samples of *P. h. capitis* were collected from school children, whose ages ranged from 5 to 14 years old, from the Arica and Parinacota region. Informed consent was obtained from parents and children. The specimens collected were maintained in a 70% alcohol solution. Only complete specimens were selected for this study (e.g. specimens with the head, thorax, abdomen and the first pair of claws).

Table 1
Adult head lice, archaeological specimens: female versus male claws dimensions.

		Tibio-tarsal claw length (μm)		Tibio-tarsal claw width (μm)		Tarsal length (μm)		n
		Left	Right	Left	Right	Left	Right	
Archaeological female	Mean	372.5	388.8	132	142.3	166.5	175.1	6
	Maximum	442.9	489.4	176.2	181.7	232.3	224.7	
	Minimum	320.6	347.4	105.6	115.2	128.3	159.4	
	SD	42.9	52.9	26.1	22.5	36.2	25.4	
Probability		p = 0.09	p = 0.9	p = 0.003	p < 0.001	p = 0.04	p = 0.5	
Archaeological male	Mean	412.7	393	183.2	197.5	205.3	184.7	8
	Maximum	449.1	422.2	218.3	217.9	216.1	206.6	
	Minimum	359.1	334.9	145.5	168.5	191.9	118.6	
	SD	32.6	26.9	25.2	14.9	10.2	28.2	

Table 2
Adult head lice, modern specimens: female versus male claws dimensions.

		Tibio-tarsal claw length (μm)		Tibio-tarsal claw width (μm)		Tarsal length (μm)		n
		Left	Right	Left	Right	Left	Right	
Modern female	Mean	342.6	340.7	121.4	124	168.1	163	13
	Maximum	385.7	394.2	149.7	152.9	206.3	186.3	
	Minimum	261.6	273.5	91.1	99.8	143.2	121.1	
	SD	34.6	34.3	15.4	13.2	17.9	21.1	
Probability		p = 0.5	p = 0.7	p < 0.001	p < 0.001	p = 0.3	p = 0.3	
Modern male	Mean	332	334.6	160.3	163.4	176.7	174.6	9
	Maximum	382.2	381.4	173.9	183	198.6	195.6	
	Minimum	273.1	250.7	150.7	152.1	137.6	111.6	
	SD	32.4	39.8	9.4	11.3	18.7	24.8	

Table 3
Adult head lice. Comparison of claws dimensions, same sex specimens by period (probability).

Sex	Sample period	Tibio-tarsal claw length (μm)		Tibio-tarsal claw width (μm)		Tarsal length (μm)	
		Left	Right	Left	Right	Left	Right
Female	Archaeological vs Modern	0.1	0.08	0.3	0.1	0.9	0.3
Male	Archaeological vs modern	0.0001	0.003	0.04	0.0001	0.001	0.4

The samples were morphoscopically analyzed at the Bioarchaeology Laboratory in the Advanced Research Institute of the Tarapacá University. The specimens were mounted on aluminum stubs, with double carbon contact face and analyzed using a Zeiss scanning electron microscope (SEM), EVO LS-10 model. We used variable pressure mode (VP), a chamber pressure of 150 Pa (low vacuum), and a pressure of 2×10^{-5} Pa (high vacuum) in the column. The working distance (WD) was 4.5–7 mm with a 0° tilt. The images of the specimens were taken at 3024×2304 pixel resolution and scanned at a speed of 12 min and 54 s. Using the SmartSem software, three variables were quantified in each specimen: A) tibio-tarsal length, B) tibio-tarsal width, and C) tarsal length for both right and left claw for the first pair of legs (Fig. 2).

The results were tabulated and analyzed with Minitab 16 and Excel software. Basic statistics and t Student tests were used to assess the hypothesis. The variation was also analyzed using box plots.

3. Results

3.1. Comparison between different sexes by period

The results of the statistical analyses indicate that the archaeological tibio-tarsal length of male was significantly greater than females ($p=0.09$), except for the right side (Table 1). In the modern sample, there were no significant differences between male and female specimens for the tibio-tarsal length (Table 2). The male tibio-tarsal width (right and left) was significantly greater than females, in both archaeological and modern samples ($p \leq 0.01$). Regarding the tarsal length, we only found differences between the left segment of archaeological male and female specimens, whereas the left tarsus was significantly larger in males ($p=0.04$). These differences indicate that the claws of male specimens, either archaeological or modern, were larger than females.

3.2. Comparison between specimens of the same sex by period

Comparing measurements of archaeological females versus modern females no statistical differences were found for any of the analyzed claws parameters, $p > 0.05$ (Table 3, Fig. 3). In contrast, comparing archaeological males versus modern males (Table 3, Fig. 4), it was found that the right and left length of tibio-tarsal claw, right and left width of tibio-tarsal claw and the left tarsus differ significantly ($p \leq 0.04$). However this variation was not observed along the right tarsus.

4. Discussion

Despite the small sample size, our results indicate that there is a significant difference between the claw size of ancient male specimens and modern lice, with the former having larger and wider claws (Figs. 5 and 6). Is this an adaptation directly related to the thickness of hair? Modern studies of hair thickness reveal a diameter of $74.5\text{--}96.1 \mu\text{m}$ (Wortmann and Schwan-Jonczyk, 2006). Mansilla et al. (2011) report values for hair diameter of $87 \mu\text{m}$ in contemporary samples (Mexican) and a range of $70\text{--}92.5 \mu\text{m}$ in Mexican pre-Columbian mummies. On the other hand, our

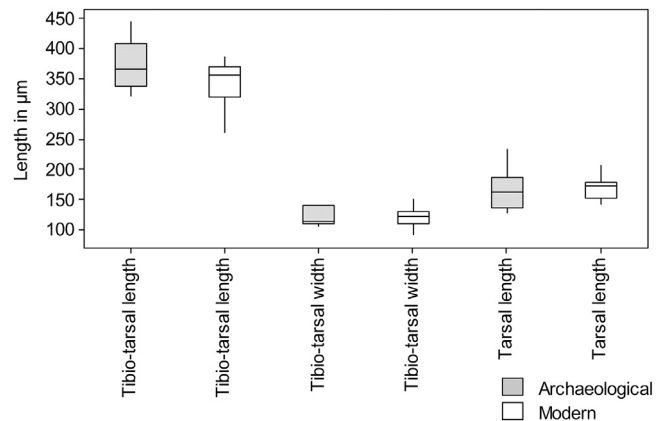


Fig. 3. Adult head lice, female specimens. Size of the left claws, archaeological versus modern cases.

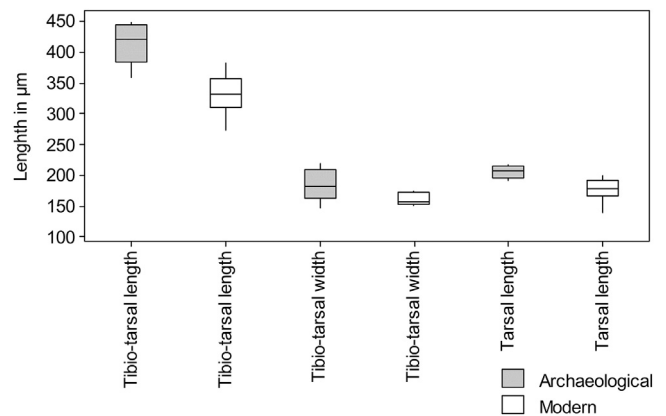


Fig. 4. Adult head lice, male specimens. Size of the left claws, archaeological versus modern cases.



Fig. 5. Claw of an archaeological male specimen of *P. h. capitis* (Case: Quiani-7 Tomb 9, specimen 8).



Fig. 6. Claws of a modern specimen of *P. h. capitis*.

hair metric studies in 21 adult pre-Columbian mummies of Arica resulted in an average of 85.7 μm (with a range of 60.5–119 μm). Our mean value fits well with those reported by Mansilla et al. (2011).

Returning to the claw morphology, Retana-Salazar and Ramírez-Morales (2006) have pointed out that the morphology of the clutches of *P. h. capitis* is directly related to the substrate to which they are affixed, and even the legs are taxonomic characters used to segregate species or to explore possible metric differences between modern *P. h. capitis* and 3000 years old specimens. However, in the female sample that we analyzed we did not find significant morphological changes through time in the structures studied. It would be expected *a priori* that both sexes should present a similar morphological reduction of their anatomy, assuming they adapted to hair thickness.

Thus, this morphological study raises a few questions: Has hair been thinning over time? If so, has the male louse lost firmness to attach itself to the female which would lead to fewer opportunities for intercourse?

The data presented for hair diameters, though small, suggests that human hair thickness has not changed much over the last few thousand years. However, we suggest that future studies should be pursued on these topics using a larger comparative study of archaeological versus modern human headlice specimens. This would allow an in-depth study of the micro-structures of *P. h. capitis* and its substrate (hair), and would open new fields of study within archaeoentomology.

Contrary to our expectations, the adult stages for archaeological specimens of *P. h. capitis* are found in an excellent state of preservation. Their presence was common in prehistoric times, as have been reported in northern Chile mummies, beginning as early as 5000 years B.C. Ancient populations also exhibited a high degree of infestation by nits (Arriaza et al., 2013).

Scanning electron microscopy contributes significantly to the study of ectoparasites affecting ancient South American populations. In our case, the metric analyses demonstrated that *P. h. capitis* has experienced morphometric changes over time, possibly adaptive, and in direct relationship to substrate gripping (hair) and the evolution of the host (Dutra et al., 2014). Likewise, Retana-Salazar and Rodríguez-Arrieta (2016) has shown that there are important variation in the micro-morphological segments of the genitalia in New World head lice population.

5. Conclusions

Our data on the first pair of claws suggest that modern *P. h. capitis* specimens are less sexually dimorphic than ancient ones. Archaeological specimens presented higher sexual dimorphism, with males having wider and larger claws. Regarding morphometric changes through time, modern male head lice have smaller claws than archaeological males. Despite the decrease in claw size over time, sexual dimorphism in the claws was present in both set of samples. Thus, it is unlikely to have affected the modern male louse's grasping capability during mating.

We suggest the hypothesis that cultural activities such as nit picking and the invention and use of delousing combs could have acted against the reproduction of larger lice specimens. That is, specimens with larger claws and overall body size could be easily picked out and/or get trapped in the teeth of the combs, generating a reduction in the size of the specimens. The data presented here are preliminary, but we hope in the future to increase the number of samples studied to give greater validity to statistical analyses and comparisons covering a wider time sequence.

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