



MOLECULAR STUDY OF HUMAN HEAD LICE, *PEDICULUS HUMANUS CAPITIS* COMPARISON WITH GOAT SUCKING LICE, *LINOGNATHUS STENOPSIS* (BURMEISTER) STRAINS

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Abstract

In this study, only (122) out of (915) primary school students were shown to be infected with head lice *Pediculus. humanus capitis*. The number and percentage of infected males were 46 (11.3%), while the number and percentage of infected females were 76 (14.9%). The results in our study also showed that the number and percentage of goats infected with goat sucking lice, *Linognathus stenopsis* was 70 (21.7%) of the total 322 animals, with the highest number and percentage among female goats 44 (62.9%) compared to the male goats 26 (37.1%). The study demonstrated that the rate of genetic difference between the studied samples was 89% and the similarity rate was 11%. Detection of OP-K01 gene pieces by PCR products showed that the amplicon size was 520 bp for *P. humanus capitis* isolated from humans, while the detection of OP-E20 and OP-M05 gene pieces with PCR product showed the lowest amplicon size 230 bp for *Linognathus stenopsis* isolated from goats.

Keywords: Molecular, *Pediculus. Humanus Capitis*, *Linognathus Stenopsis*, Genotypes.

دراسة جزيئية لقمل الرأس في الإنسان *Pediculus. humanus capitis* مقارنة مع

قمل الماعز الماص

Linognathus stenopsis (Burmeister) strain

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الملخص

في هذه الدراسة تبين إصابة 122 طالب بقمل الرأس *Pediculus. humanus capitis* من مجموع 915 من طلاب المدارس الابتدائية. حيث كان عدد الذكور المصابين 46 بنسبة 11,3% بينما عدد الاناث المصابة 76 بنسبة 14,9%. كما بينت نتائج دراستنا بأن عدد حيوانات الماعز المصابة بقمل الماعز الماص *Linognathus stenopsis* 70 بنسبة 21,7% من مجموع 322 حيوان، حيث كانت الإصابة بين الاناث اعلى 44 بسنة 62,9% مما عليه بين الذكور 26 بنسبة 37,1%. أظهرت الدراسة أن معدل الاختلاف الوراثي بين العينات المدروسة كان 89% وكان معدل التشابه 11%. اظهر تفاعل البوليمراز المتسلسل PCR بان حجم التضاعف باستخدام البادئ OP-K01 كان (520 bp) بالنسبة لعينات قمل الرأس *P. humanus capitis* المعزولة من الانسان، في حين بين كل من OP-E20 و OP-M05 اقل حجم للتضاعف (230 bp) بالنسبة لعينات القمل *Linognathus stenopsis* المعزولة من الماعز.

كلمات مفتاحية: جزيئية، قمل الرأس، الانسان.

Introduction

Many species of endoparasites such as blood flukes, tapeworms, hookworms and ectoparasites such as fleas, mites and lice range from being annoying to human to vector-borne diseases such as bubonic plague and other diseases which have contributed to large numbers of deaths (13). Lice are ectoparasites that play the role of vectors which are able to transmit typhus infection through their bites(6).They were traditionally categorized into two orders, Anoplura (sucking lice) and Mallophaga (chewing lice) (15 and 12). *Pediculus humanus capitis* known as head louse is an obligatory hematophagous ectoparasite of humans belonging to the order Anoplura, Family Pediculidae. Its life cycle includes eggs that hatch in 5-10 days, three nymphal instars and adults which live about 30 days depending on appropriate temperature and

provided blood meals. The life cycle takes from 16-21 days (1). *Linognathus stenopsis* is a domestic goat blood sucking louse belongs to the Order Mallophaga, Family Linognathidae, and its life cycle takes 2-3 weeks in good conditions (11). *L. stenopsis* is transmitted from one goat to another by rubbing and body contact. Goat sucking lice are generally introduced into the herd by bringing infested goats from outside. A number of sucking and biting lice begin to increase during the fall season and populations reach peak in early spring. Summer populations are usually low infested animals and may suffer from weight loss, anemia and reducing of milk production (10,9). The aim of this study is to determine the genetic strains between the *P. humanus capitis* ectoparasites and animal ectoparasites strains and their effects on human's health. Random Amplified Polymorphic DNA (RAPD) is a technique of polymerase chain reaction which can be used to generate DNA fragments which can further help to estimate the genetic diversity of any organism. These RAPD markers can aid in estimating and measuring genetic differences between various individuals, species or even populations (5).

Materials and Methods

A total of 915 randomly student samples were selected from five primary schools for boys and girls in AL-Door city/south of Tikrit during the period from 1st December 2017 to 15th March 2018. Direct optical hair examination for louse (adult and/or ova) was done by a nursing inspector and other information {sex, family size, hair length, age (years) and mother's employment} were collected from the infected individuals and the school principal. The study also included 322 goats during December 2017. It was observed that the goat ectoparasite was present in abdomen, back, the space below the shoulders, around the teats (females) and scrotum (males) of the infested animal's body. The collection of lice was made during many visits per week, and 10 lice were taken at each visit. The collected lice were transferred to the laboratory and examined by using stereomicroscope for specie identification depending on morphological characteristics and taxonomic keys of the parasite. The genomic DNA of *P. humanus capitis* and *L. stenopsis* were extracted following the instructions of a G-spin DNA extraction kit (intron biotechnology/Korea). The RAPD-PCR amplification by ten primers were used in this study: OP-C05GATGACCGCC, OP-E20AACGGTGACC, OP-H01GGTCGGAGAA, OP-I02GGAGGAGAGG, OP-K01CATTCGAGCC, OP-L05ACGCAGGCAC, OP-L20TGGTGGACCA, OP-M05GGGAACGTGT, OP-M06CTGGGCAACT, OP-M14AGGGTCGTTC.

Amplifications were done by Maxime PCR Pre Mix kit (i-Taq) (Intron / Korea) consisted of 5U/ μ l of i-Taq DNA Polymerase, 2.5mM of DNTPs, 1X of reaction buffer (10X) and 1X of Gel loading buffer. The specific interaction mixture of gene diagnosis consisted of 5 μ l of Taq PCR Pre Mix, 10 picomols/ μ l of primer, 1.5 μ l of DNA, 17.5 μ l of distilled water and 25 μ l final volume. The perfect conditions of gene detection were: Initial denaturation at 95 C⁰ for 3 min. (No. of cycles = 40), Denaturation was -2 at 95 C⁰ for 1 min. (No. of cycle= 40), Annealing at 35C⁰ for 1 min. (No. of cycles = 40), extension was -1 at 72C⁰ for 1min. (No. of cycles = 40) and extension was -2 at 72C⁰ for 10 min. (No. of cycles = 40).

Statistical analysis: The statistical analysis of the results was performed using an ANOVA test to demonstrate the genetically similarity between the two species under study.

Results and Discussion

Prevalence of *P. humanus capitis* among individuals in relation to the selected characteristics. Table 1 showed that the head lice, *P. humanus capitis* varied in intensity between males and females. The percentage of female infection was the highest 14.9% compared to males 11.3%. The age years ranging from 9-10 showed the highest percentage of infection i.e. 15.9%. The percentage of infection in families having members more than 6 is higher 13.6% than the families with number of members less than 5 i.e. 12.3%. The mothers that are not employed outside the house showed a greater 13.6% percentage of infection.

Table 1 Prevalence of *P. humanus capitis* among pupils in relation to selected characteristics.

characteristic	Total examined No.	Positive pupils		p-value	
		No.	%		
Sex	Male	406	46	11.3	0.111 ^{N.S}
	Female	509	76	14.9	
Age years	6-8	349	44	12.6	0.533 ^{N.S}
	9-10	176	28	15.9	
	11-12	390	50	12.8	
Hair length	Long	404	66	16.3	0.017 ^{**}
	Short	511	56	10.9	
Mother's employment	Employed outside the home	174	21	12.1	0.586 ^{N.S}
	Not employed outside the home	741	101	13.6	

Family size (No. of members)	5 ≤	204	25	12.3	0.607 ^{N.S}
	6 ≥	711	97	13.6	

** means significant differences, N.S means Non significant differences.

Prevalence of *L. stenopsis* among goats Table 2 showed that out of the total 322 animals, 70 (21.7%) were infected with goat sucking louse *L. stenopsis*, and the highest number and percentage of infections was among female goats 44 (62.9%) in comparison with the number and percentage of the infected male goats 26 (37.1%). This is due to a number of reasons, including physiological related to the size of females compared to males.

Table 2 Prevalence of *L. stenopsis* among goats

Total examined No.	Positive goats		Male		Female		p-value
	No.	%	No.	%	No.	%	
322	70	21.7	26	37.1	44	62.9	0.981 ^{N.S}

N.S means Non significant differences

RAPD-PCR amplification of *P. humanus capitis* and *L. stenopsis*. The PCR product size pieces of OP-K01 gene of *P. humanus capitis* and *L. stenopsis* by using the DNA extraction showed that the ladder 100 bp and amplicon size was 520 bp for samples isolated from humans, while the products of OP-E20 and OP-genes showed that the amplicon size was 230 bp for samples isolated from goats as shown in figure 1.)"

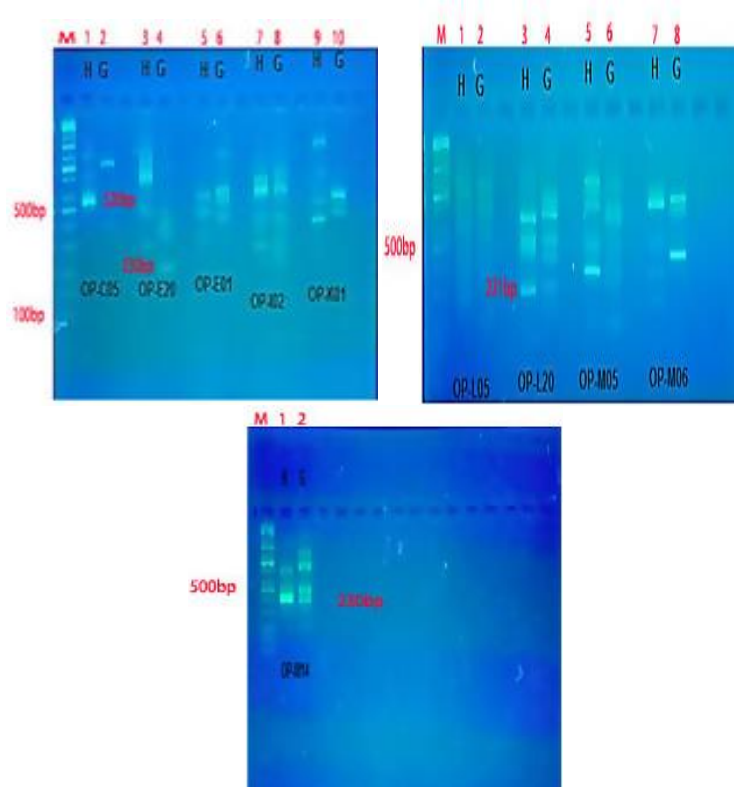


Figure 1 RAPD patterns of *P. humanus capitis* of human and *L. stenopsis* of goats with ten pieces of genes OP-C05, OP-E20 , OP-E01 , OP-I02 , OP-K01 , OP-L05 , OP-L20 , OP-M05 , OP-M06 and OP-M14.

Pediculus humanus capitis is a serious parasite that infects humans and transmits serious skin diseases as well as typhus fever (7). The similarity of genes between the two different species *P. humanus capitis* and *Linognathus stenopsis* may help the parasite to be transferred from animals to humans. Our findings agreed with (2) reported the genetic structure of human louse populations by analyzing the nuclear genetic variation at 15 newly developed microsatellite loci in 93 human lice from 11 sites in four world regions (2). The spread of this parasite among children, especially school children, was especially fortunate among pupils in rural schools suffering from lack of hygiene and care for these ages of children. These results were in a harmony with (8) who found that the children are more likely to have this parasite with head lice due to lack of health awareness, lack of hygiene and sterilization (8). The amplified size pieces of OP-K01 gene of *P. humanus capitis* and *L. stenopsis*, in both humans and animals are very genetically similar. There may be a joint infection of humans due to cohabitation with animals, especially among those that breed cattle. There may be genetic mutations of genes in these external parasites, causing serious pathogenesis, especially in disease transmission, and this matches with what has been found by Drali (4) stated that the 908-bp DNA fragment of the VSSC gene, encoding the subunit of the sodium channel and encompassing the three mutation sites, was PCR sequenced from 65 lice collected from a homeless population. We observed a high prevalence of the 3 indicated mutations in the body of lice collected from homeless people (4). Genetic mutations of some of the head lice strains may lead to the emergence of a strain that may be more severe that makes the parasite to have more impact on human's head scalp and transmission of disease. These results corresponded with (14) showed that the genetic mutation of *humanus capitis* leads to a severe disease transmission because they are associated with high incidences of diseases and high mortality rates, particularly epidemic relapsing fever and typhus, which could be fatal in up to 40% of patients (14). The genetic similarity between head lice and goat lice suggests that there is a direct relationship between these two parasites, and that some of them may move from goats to humans because of coexistence, especially in farms and fields in rural areas. These findings agreed with Boutellis (3) reported that differences between head lice, body lice and some animal

lice are a genetic shift subject to environmental conditions and a change may occur in some inherited traits (3).

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