Comparing the Efficacy of Commercially Available Insecticide and Dimeticone based Solutions on Head Lice, *Pediculus capitis: in vitro* Trials

Piyasada Bulunan İnsektisit ve Dimetikon Bazlı Baş Biti Ürünlerinin Etkinliklerinin Karşılaştırılması: *in vitro* Denemeler

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ABSTRACT

Objective: Head lice infestation is a public health and social problem for almost all countries worldwide. For its treatment, insecticide and dimeticone-based solutions are currently available in the markets in many countries. We aimed to compare the efficacy of commercially available anti-head lice shampoos containing insecticide and physically effective products with different percentages of dimeticone using an *in vitro* technique.
Methods: Head lice specimens were collected from primary school children using special plastic and metal combs. Anti-head lice products were commercially purchased and used directly. The specimens were placed one by one in 5-cm Petri dishes containing a slightly wet filter paper and were kept in a plastic cage at 28±2°C and 50%±20% relative humidity. A standardized protocol was used for testing all the products, and mortality data were obtained after 24 h. Two control tests were performed with each batch of trials. For each product and control, 10-20 head lice specimens were used, and the results were statistically analyzed.
Results: Our study demonstrated that among all the tested products, two products containing mineral oils [5.5% dimeticone & silicone (patented product) and dimeticone (no percentage mentioned in the prospectus) & cyclopentasiloxane] were found to be more effective for killing head lice *in vitro*.
Conclusion: Physically effective products can be repetitively used because they are non-toxic and resistance to them is not expected. To control the infestation at a public level, the use of these products needs to be encouraged with respect to their cost price.
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Keywords: Head lice, dimeticone, synthetic insecticides, efficacy, *in vitro* testing

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ÖZ

Amaç: Baş biti enfestasyonu tüm dünyada bir halk sağlığı ve sosyal problemdir. Tedavisi için insektisit bazlı ve dimetikon bazlı solüsyonlar kullanılmakta ve günümüzde birçok ülkede bulunabilmektedir. Bu çalışmada piyasada bulunan insektisit bazlı baş biti ürünleri ile farklı oranlarda dimetikon içeren fiziksel olarak etkili ürünlerin etkinliklerinin *in vitro* olarak karşılaştırılması amaçlanmıştır.
Yöntemler: Baş biti örnekleri, özel taraklar yardımıyla ilkokul öğrencilerinden toplanmıştır. Ürünler ise piyasada bulunduğu şekli ile satın alınmış ve kullanılmıştır. Örnekler, içinde hafifçe ıslatılmış filter kağıdı bulunan 5 cm petri kaplarına tek tek konulmuş ve 28±2 °C ile %50±20 nem sağlanan ortamda tutulmuşlardır. Standardize edilen protokol bütün ürünler için aynı şekilde uygulanmış ve 24 saat sonra ölüm oranları not edilmiştir. Kontrol testlerinde örneklere herhangi bir şey uygulanmamıştır. Her ürün ve kontroller için 10-20 baş biti örneği kullanılmış ve sonuçlar istatistiksel olarak analiz edilmiştir.
Bulgular: Çalışmamız mineral yağ içeren iki ürünün (%5,5 dimetikon ve silikon ile dimetikon (oranı prospektüste verilmemiş) ve siklopentasiloksan) baş bitlerinin öldürülmesinde daha etkili olduğunu göstermiştir.
Sonuç: Toksik olmamaları ve herhangi bir direnç gelimesi beklenmediği için dimetikon bazlı ürünler tekrarlanarak kullanılabilir, ancak enfestasyonun halk sağlığı düzeyinde kontrol altına alınabilmesi için fiyat açısından rekabet edebilecek düzeye getirilerek bu ürünlerin kullanımının teşvik edilmesi gereklidir. *(Turkiye Parazitol Derg 2015; 39: 305-9)*

Anahtar Kelimeler: Baş biti, tedavi, dimetikon, sentetik insektisitler, etkinlik, *in vitro* testler

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INTRODUCTION

Head lice infestation that is caused by *Pediculus capitis,* a blood-feeding ectoparasite living in human hair, is a public health and social problem for almost all countries, particularly developed ones. Different countries have reported infestation rates of between 1.8% and 87% (1). Although treatment using various commercial preparations appears to be easy, re-infestation is very common because of untreated individuals in the population and resistance acquired against commonly used chemical insecticides. Because of the spread of head lice by head-to-head contact and sharing hair-contacted materials such as hair-pins, infestation is more common in schools and community-dwelling units. The infestation rate is always higher in girls than in boys because their hair is longer and they share hair accessories (2).

Head lice are obligate ectoparasites of humans that live on human scalp and can survive only on human blood. Both male and female head lice begin feeding on their host as soon as a nit hatches; adult females may lay up to 300 eggs in their life span. Because they are hemimetabolous insects, three life stages are observed in their life cycle: egg (nit), nymph (immature form after hatching and feeding on blood), and adult. It requires approximately 15-20 days for an egg to develop into an adult, and adults live for approximately 30 days (2). Besides causing scalp irritation and discomfort, pediculosis also causes social ostracism, anxiety in parents, and absence at school, even with light infestations (3).

In Turkey, a total of 31 studies have been conducted so far, mostly among school children, and the average infestation rate was reported to be 10.16% (min, 0.54%; max, 29.4%). All the examinations were performed by eye inspection, and girls showed >6 times higher infestation rate than boys (4).

Insecticide-based anti-head lice products have been used since a long time worldwide; however, resistance against synthetic chemical insecticides, such as synthetic pyrethroids, is becoming more prevalent (5-8). Moreover, pesticide-based products are toxic for children and its long-term effects on human health still remain unclear. Therefore, physically effective products containing dimeticone and some other ingredients that are not harmful to human have been developed as alternative treatments, and they are currently available in the markets in many countries. In Turkey, insecticide-based and physically effective products are currently available in the market. Because resistance is not possible, physically effective products are being suggested in recent years. However, price discrepancies are in favor of insecticide-based anti-head lice products rather than physically effective products.

Because infestation is very common worldwide, the market of anti-head lice products is very big and is economically important. This study aimed to compare the efficacy of commercially available anti-head lice products containing insecticide and physically effective products containing different percentages of dimeticone derivatives by an *in vitro* technique.

METHODS

Anti-head lice products and *P. capitis* specimens

All anti-head lice products were commercially obtained and were used directly, and tests were applied as a single-blind study. Three commercial, synthetic insecticide-based shampoos [A: 0.4% Sumithrin; B: 0.6% permethrin; and C: 0.3% pyrethrin & 3% piperonyl butoxide] and three physically effective products containing different percentages of dimeticone derivatives [D: 5.5% dimeticone & silicone; E: mineral oils & foaming agents; and F: dimeticone (no percentage mentioned in the prospectus) & cyclopentasiloxane] were used for the *in vitro* trials. The instructions provided by the producers within the product boxes (prospectuses) were used to obtain information regarding the ingredients of the products; the manufacturers were not contacted.

Children from a village primary school were screened for the presence of head lice after obtaining ethical permission from the Celal Bayar University Faculty of Medicine, Ethics Committee (no: 2015-107) and written informed consent from parents or teachers of the school children. The study protocol was also approved by the director of the school. Head lice specimens, both adults and nymphs, were collected from infested children using special plastic and metal combs.

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In vitro trial

For accurate comparison, a standardized protocol was used for testing all the products. In brief, the head louse were covered with 20 microliters of the product for 20 min and were then washed using 100 ml of tap water to eliminate product residue. The specimen were carefully transferred to a new 5-cm petri dish containing slightly wet circular-shaped filter papers and several undyed hairs to prevent desiccation of the specimen. Head lice were individually observed under a stereomicroscope at 5, 10, 15, 20, 30, 60, 120, 180, and 360 min and after 12 and 24 h. Each specimen was categorized according to their vital signs as follows: physical movement (inability to walk and antenna/claw/leg twitch), internal organ function (stomach musculature contractions), and their response when the legs were stroked with a forceps (9-12). For the control group, the head lice were directly placed in petri dishes without any treatment. For each specimen, if active movement was observed in the outer or internal organs, the louse was considered "alive;" if no movements were observed, the louse was considered "dead." At least two control tests were performed with each batch of trials. For each product and control batch, 10 head lice specimens were used.

Statistical methods

Analysis of variance was used for statistical analysis. A p value of <0.05 was accepted as significant. In addition, Bonferroni test was used for post-hoc analysis.

RESULTS

The results were evaluated with respect to two categories. (i) Mortality rate after 24 h: among synthetic insecticide-based shampoos, product C, and among physically effective products, products D and F, killed all specimens at the end of 24 h; howev-

er, product D was faster effective one. Product D started to kill the head lice in 5 min, with a 60% mortality rate, whereas other products reached a 60% mortality rate between 15 and 360 min (Figure 1). After 24 h, the mortality rate of products C, D, and F-treated head lice was statistically different from the control group (p<0.0001). (ii) The average duration for killing head lice was 88.5 min for product F; 99.5 min for product D, and 110.7 min for product C (Table 1). These three products were statistically compared with each other, and the differences were not found to be statistically significant for the average duration of killing efficacy (p=1.00).

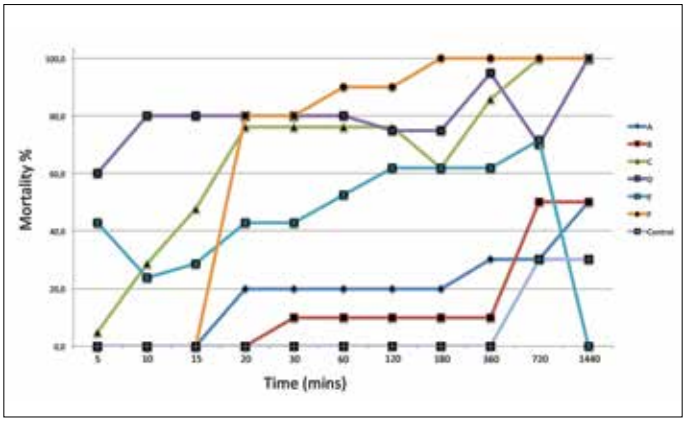

Figure 1. Average mortality rates of treated head lice specimens with respect to time

DISCUSSION

The results of this *in vitro* trial clearly demonstrated that the products D (containing 5.5% dimeticone & silicone) and F (containing unknown dimeticone & cyclopentasiloxane) have shortest duration for killing head lice among all the tested products. Regarding product F, we could expect its good efficacy because it contained both silicon oils, although no information about their percentages was there in the prospectuses. The high efficacy of product D can be compared with another product containing 4% dimeticone, which is one of the commonly used products in Europe (13). Additionally, the product C containing 0.3% pyrethrin & 3% piperonyl butoxide was found to be more effective among the three insecticide-based shampoos.

The cure rate was reported as approximately 70% after the appli-

Table 1. Average duration for killing head lice according to the products

		Products	N	Mean (min)	95% Confidence Interval for Mean		Minimum (mins)	Maximum (mins)	p
					Lower Bound	Upper Bound			
Outer organs*	SIP	A	10	914.0	487.8	1340.2	20	1440	p<0.0001
		B	10	867.0	429.2	1304.8	30	1440	
		C	21	110.7	52.8	168.6	5	360	
	PEP	D	20	99.5	26.3	172.7	5	360	
		E	21	479.0	190.5	767.6	5	1440	
		F	10	88.5	-70.3	247.3	15	720	
		Control	10	828.0	505.7	1150.3	360	1440	
Internal organ**	SIP	A	10	843.5	371.6	1315.4	15	1440	p<0.0001
		B	10	865.5	426.1	1304.9	15	1440	
		C	21	107.9	50.4	165.3	5	360	
	PEP	D	20	40.5	-10.6	91.6	5	360	
		E	21	440.0	143.0	737.0	5	1440	
		F	10	5.0	5.0	5.0	5	5	
		Control	10	828.0	505.7	1150.3	360	1440	
Final death	SIP	A	10	914.0	487.8	1340.2	20	1440	p<0.0001
		B	10	867.0	429.2	1304.8	30	1440	
		C	21	110.7	52.8	168.6	5	360	
	PEP	D	20	99.5	26.3	172.7	5	360	
		E	21	479.0	190.5	767.6	5	1440	
		F	10	88.5	-70.3	247.3	15	720	
		Control	10	828.0	505.7	1150.3	360	1440	

* antennas, claws and legs; ** stomach musculature contractions; min: minutes; SIP: synthetic insecticides; PEP: physically effective products

cation of the anti-head lice products containing 4% dimeticone (14). In natural applications, it is always better to keep the lotion for extended periods to prolong the contact time. In this study, the specimens were immersed for only 20 min and were then immediately washed. The contact times indicated in the prospectuses could not be applied in this study, and this could be the reason for the differences in the mortality rates of the products. The lethal efficacy of the products could also change in longer applications; however, we believe that a standard protocol needs to be used for *in vitro* trials, as we did in the study.

The physically effective products for head lice treatment are currently widely used and are considered non-toxic when topically applied. Studies testing their efficacy always emphasize that resistance will not develop (15). These products contain different percentages of dimeticone together with some different accessory ingredients that can be useful for enhancing the efficacy of anti-head lice products or for cosmetic reasons. As known, dimeticones are silicon oils and are commonly used in cosmetic products. Its mode of action to kill head lice is very simple; it penetrates into the spiracles laying both sides of the abdomen of lice. Spiracles are the breathing pores of lice and are also where the lice eliminate water after blood sucking. Blocking the spiracles discontinues airflow (asphyxia) and water, leading to death (16, 17).

The main target of the chemical products or synthetic insecticides is the nervous system of the lice. Pyrethrins are derived from a natural compound, chrysanthemum extract, and their mode of action is interfering with the sodium transport in lice. This causes neurotoxicity and eventual paralysis. Some products contain piperonyl butoxide that has a synergistic effect when combined with pyrethrins (18, 19). This compound is being used in many insecticide formulations as anti-mosquitoes agents (20). In this study, only product C, containing piperonyl butoxide, had a high and fastest efficacy among the three insecticide-based products tested. However, according to the prospectus of product C, hair needs to be washed after 10 min probably because of the possibility of the chemical compound being absorbed, thus decreasing its *in vivo* efficacy.

After applying the products and washing of the specimens, the monitoring continued until 24 hours, as suggested in previous studies (5, 15), related to the recovery of head lice over time. In this study, 70% of control lice were alive after 24 h, but movements in the external or internal organs were observed in some lice specimens after 12 and 24 h in some products, and thus, we are agree that specimens should be kept for over 24 h to obtain mortality data, with an effort to prevent desiccation.

CONCLUSION

Our study demonstrated that among all the tested products, products D and F, containing silicon oils, were more effective in killing head lice *in vitro*. The physically effective products can be repetitively used because they are non-toxic and resistance is not expected to develop. To control head lice infestation at a public level, the use of these products needs to be encouraged with respect to their cost price.

Ethics Committee Approval: Ethics Committee approval was received from Celal Bayar University Faculty of Medicine, Ethics Committee (no: 2015-107) for this study.

Informed Consent: The written informed consent from parents or teachers of the school children were obtained in the study.

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