The Prevalence of Head Lice and Health Consequence of Their Medical Treatments among Female Students from Different Schools

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ABSTRACT
Background and Objectives: Head lice infestation becomes a pressing problem around the world. This study characterized the prevalence of head lice and health consequences associated with their medical treatment among female students.

Materials and Methods: A cluster of five schools was selected randomly in nine locations in Gaza Strip. The abundance of head lice among students was determined manually using a special crown. Each female student was required to comb her hair five times over a white sheet of A4 paper. Each sheet was collected and coded by the name of the student and kept in covered plastic cups. The samples were transferred to the lab for analysis. The second part of the study included voluntarily collection of a 5 mL blood and urine sample.

Results: The highest prevalence of head lice (18%) among primary schools in the middle zone cluster, followed by Bait Lahia and Khan Younis clusters, whereas the lowest prevalence was found in the Gaza cluster (7.14%). Blood analysis showed an elevation of aspartate aminotransferase (AST) and urea levels and a reduction in acetylcholinesterase (AChE) activity. Pesticide residue analysis showed positive values in head lice patients who intensively used medical treatments.

Conclusion: The results of this study showed that head lice medical treatment has health consequences.

Key words: head lice, Nymph, eggs, Health consequences, Density of insects, Medical treatment.

INTRODUCTION
Pediculosis capitis, or head lice, is a blood-sucking insect of public health importance and a specific parasite of human beings. Pediculus humanus var capitis lives on the head (head lice).

The female insect lives for 4-12 weeks, lays approximately 250-350 eggs and dies when separated from the host. The insect is usually found on the heads of young boys and girls. This insect creates many health problems. The distribution of head lice between populations may occur through personal clothing exchange, towel sharing, hairbrush sharing, and direct head-to-head contact.

It is an obligate ectoparasite insect that creates frequent public health problems\textsuperscript{1, 2}. Head lice infestation is usually detected by three modes: inflammation of the scalp and neck, sighting of lice, and detection of eggs attached to hair shafts\textsuperscript{3}. Although it does not transmit any pathogenic agents, several authors\textsuperscript{4,6} have revealed complications derived from head lice parasitism, such as scalp lesions caused by scratching and secondary bacterial infections. The kindergarten and/or school environment is the easiest mode of pediculosis cross-transmission among boys and girls.

The prevalence of pediculosis among children aged 6–12 years has previously been reported; for instance, Alempour Salemi \textit{et al.}\textsuperscript{7} found infestations in approximately 27% of urban primary schools in Southeast Iran, whereas Hodjati \textit{et al.}\textsuperscript{8} found a lower infestation rate (3.64%) in school children of Tabriz City. In Egypt, the prevalence reached approximately 20.7% in public schools and 9.04% in private schools, and occurred 25.8% more often in girls than in boys\textsuperscript{9}. In the USA, a 25% prevalence was reported in school-aged children\textsuperscript{10}.

Several trials\textsuperscript{11,13} have been conducted to control head lice infestations among boys and girls. These included the use of insecticide shampoos. However, all of these trials may have a negative effect on the control process, such as the appearance of resistant insect populations. The resistant insect populations make pediculosis control more complicated and expose children to the toxic effects from medical treatments.

Head lice resistant to medical treatment (insecticide shampoos) may result in the use of other types of insecticides that may cause toxicity in children. The prevalence of head lice and the health consequences of medical treatment among primary school children have not been investigated in Palestine and surrounding countries, or the investigation remains in the primary stages. There are no scientific reports from Gaza on the prevalence of head lice.

The authors of this study investigated the prevalence of head lice in primary, elementary and secondary schools in the Gaza Strip, Palestine, and investigated the health consequences associated with the medical treatment of children. The study included only female students because the majority of male students in primary schools had very short hair (less than 0.5 cm) due to recent health regulations.

\section*{MATERIAL AND METHODS}

\textbf{Site description}

\textbf{Characterization of head lice infestation}

Clusters of five primary schools were selected randomly from major cities in the Gaza Strip, as shown in Table 1. Five classes from each school were randomly selected. Each female student in the class was given a special hair brush to collect adult head lice, newly hatched lice (nymphs) and eggs. In some cases, the school teacher helped the students collect head lice (Figure 1).

Each female student was given a code number and required to use the hair brush five times around her head to collect the head lice on a white sheet of A4 paper. Then, the students put their sheets in separate plastic cups with covers to prevent lice from escaping from the cop. The cups were closed and transferred to the laboratory for counting and characterization. The counts included adult lice, nymphs and eggs.

The positive cases were recorded and given a special code for blood and urine sample collection.

\textbf{Effect of student age in head lice infestation.}

The procedures described above were repeated in one elementary and one secondary school to investigate the effect of age on the prevalence of head lice infestation.

\textbf{Medical treatments}

Medical treatments included shampoos containing different active ingredients against head lice. Health department allows the use of those active ingredient to control infestation among children.

\textbf{Blood and urine collection}

Blood and urine samples were collected only from infected children with head lice. The volume of the samples was 5 mL each. The
samples were coded with the name of the student, kept in ice box for transfer to the laboratory and stored in a refrigerator for analysis the next day. Control blood and urine samples were collected from head lice-free students. The samples were collected during the academic year 2016/2017. Health records of children were collected using a self-administered questionnaire. Blood samples were used to determine AST, urea and AChE activity and urine sample to analyze the consequences of using insecticidal shampoo products.

Responses of liver biomarkers to medical treatment of head lice
The response of the liver to the head lice medical compounds was estimated by determining the activity of AST in the blood serum according to the method of Reitman and Frankel\textsuperscript{14} with a commercially available kit purchased from BioMerieux. The AST test is a blood test that checks for liver damage.

The percentage of enzyme activation was calculated according to El-Nahhal \textit{et al.}\textsuperscript{15}.

Responses of kidney biomarkers to medical treatment of head lice
The response of the kidney was estimated by determining the concentration of urea in blood serum, which is a parameter that characterizes the kidney response to the medical compound used for head lice treatment. The abovementioned responses were determined according to well-known international methods.

Response of AChE to medical treatment of head lice
AChE activity in blood serum was used as an indicator of the nervous system response to medical treatment. The activity was measured spectrophotometrically according to Ellman \textit{et al.}\textsuperscript{16}.

Determination of malathion residues in urine samples
Shampoo containing malathion was purchased from a local certified pharmacy and used to extract standard malathion material for GC determination following the procedure described previously\textsuperscript{17,18}. Extraction of malathion residues from urine samples was performed following the procedure previously described\textsuperscript{19}. The extracts were cleaned up using anhydrous sodium sulfate to ensure removal of water droplets. Then transported to crimp vials for GC determination following the procedure described previously\textsuperscript{20}. To determination of percentage recovery of malathion from urine sample, nine urine samples of 5 mL each were collected from the control group and transferred to the extraction tubes. Then, samples were subdivided into three groups with three replicates each. Group 1, 2 and group 3 received 0.1, 0.2 and 0.5 mg respectively. The samples were carefully shaken. Then, the extraction procedure described by Ref\textsuperscript{19} above was carefully followed to determine percentage recovery.

Statistical Analyses
The data for each cluster were combined, coded, and analyzed using the Excel program. Averages, standard deviations, and percentages were calculated as previously described\textsuperscript{15}. Significant differences were calculated by a t-test; p-values equal to or below 0.05 indicate significant differences.

RESULTS AND DISCUSSION
The data in Figure 1 show the tools used to manually collect the head lice, a part of the head lice life cycle directly on the head of a primary school child and a sample collected from a child head. The tools were effective in collecting lice population because crown (Figure 1) has two working directions, one to collect adult insects and the other to collect eggs and nymph. The openings between the crown teeth are approximately 0.2 and 0.063 mm; brushing in the direction of the 0.2 mm can collect the adults, and brushing in the other direction can collect nymphs and eggs. However, the effectiveness of the tools was checked by visual investigation of the heads of children presenting zero lice. In those cases, no head lice were found, indicating the effectiveness of the manual tools.

The life cycle of head lice is well described in the literature, but we have provided an in vitro practical photo of eggs hidden in hair (bottom photo left). First, 2nd,
and 3rd instar nymphs and adult males and females were found in the collected samples (bottom photo right). Furthermore, the presence of adult males and females on a single head indicates a high prevalence of infestation.

The prevalence (% infection) of insect infestations among primary school children in different clusters (Table 1) indicated variations of health measures among students and schools. Furthermore, the schools were located in three main areas, rural, agricultural or urban. This indicates different environmental health conditions among tested schools in the clusters. It seems that the environmental conditions and student hair cover play a critical role in the infestation percentage.

The explanation for the results in Table 1 is that the clusters are located in different regions, such as agricultural, rural and urban, and the populations in these areas have different income levels, which can affect the personal medical care of children. For instance, children from agricultural and rural areas are of lower incomes, and personal health care is neglected or ignored, whereas children from urban cities are of higher incomes than children from agricultural or rural areas; accordingly, more attention is paid to personal health care. This explanation is in accordance with Abd El Raheem et al., who found different prevalence levels of head lice among students from different cities in Egypt. Furthermore, similar results were found in Iran and Jordan. Moreover, children living in agricultural areas spend more time outdoors playing and gathering, as in Khan Yonis east and rural areas in the middle zone, which had the highest number of infected cases. In fact, rural and agricultural areas have many domestic animal breeding sites for animals such as rabbits, goats, chicken and pigeons. These breeding sites may become a source of lice, which can then infect student populations living very close to those sites, leading to the spread of infection among children in schools. Our results are in agreement with previous reports that revealed different prevalence rates of head lice in different cities.

Additionally, the presence of chicken houses and animal houses among densely populated areas in Gaza may positively contribute to the rapid spread of head lice. The prevalence of lice infestation among children in urban schools was lower than the prevalence among children in rural and agricultural schools. The reason behind this is similar to that given above. In addition, the children in primary schools spend a longer time outdoors playing football and other games. This activity increased the children’s exposure time to infected cases. In addition, environmental factors may enhance the transfer of insects from infected patients to non-infected ones in accordance with Burrows, who revealed transfer mechanisms as a dispersal method to increase infestation. Nevertheless, statistical analyses indicated significant differences among infected children from agricultural, rural and/or urban areas.

The prevalence ranged from 1.08-10.41 in different schools (Table 2). It appeared that the population density of adult lice, nymphs and/or eggs per head was quite different from one school to another. Moreover, correlations between infected cases and population density (average number of lice/head) indicate a strong positive association existed, as shown by the high value of the regression coefficient ($R^2=0.8716$). Furthermore, the density of the insect population per head (Table 2) showed different values among schools. The explanation of these results is that with highly infected cases, as in the schools Gaza A and Bait Lahia A, the possibility of infection is higher than with less infected cases. In addition, epidemiological factors such as school location, family size of the children, education level of the parents, and the number of hair washings per week may have a direct role in the infestation rate and insect density among children. Similar factors have previously been suggested by Shayeghi et al. in a different investigation. Statistical analysis indicated a significant difference, p-value is less than 0.05.

The effect of student age on the distribution of infection showed the lowest
percentage of infection in the secondary schools (2.74±1.3), followed by elementary schools (5.71±2.3), and the highest percentage of infection was observed in primary schools (8.5±1.2). An explanation for these results is that children at secondary schools spent less time outdoors than children at primary schools, consequently shortening the exposure time for infection. Furthermore, the children at secondary schools are nearly self-reliant and independently take care of their personal care without parents, whereas children of primary schools are not self-reliant; they depend on their parents for their personal care. This explanation is in agreement with Khamaisih, who revealed the importance of age group in the prevalence of head lice.

A comparison with the surrounding and international countries, revealed the prevalence of head lice in children from kindergarten and schools from Israel, children from schools in Egypt, children from schools in Saudi Arabia, children from schools in Iran, children from Jordan, children from the UK, USA, and Germany. However, the prevalence among international and/or surrounding countries is quite different due to different educational levels, medical care, weather and living conditions. The detection of head lice in children around the world indicates wide spread of insect.

Medical treatments of infected students

The common name, active ingredient, lethal dose required to kill 50% of lice (LD50), and frequency of medical treatment are shown in Table 3. It was found that shampoos contained different types of insecticides and different concentrations.

The compounds used in the formulation of medical treatments were permethrin, malathion, piperonyl butoxide and isododecane (Table 3).

The use of these shampoos has been associated with health consequences such as biochemical changes in the body of infected patients (IP) and malathion residues in urine samples (Table 2). Elevated levels of AST among infected students were found in many schools; the highest level (71%) was found in Bait Lahia C, whereas the lowest level (2%) was found in Bait Lahia B. Additionally, elevated levels of urea were found in many cases, indicating impairment of kidney function. A considerable reduction in AChE was found among children who used the medical treatments compared with the control group. In addition, malathion was also detected in some urine samples, indicating a potential health hazards among students. A strong positive correlation was found between elevated levels of AST and the frequency of medical treatments, whereas the correlation was weak with urea or AChE. additionally % recovery of malathion from the control group was 85 and the standard curve was linear in the range of 0.1-6 mg/L with a detection limit of 0.01 mg/L. The explanation of these results is that the compounds (Table 3) are toxic insecticides widely used in the agricultural sector for insect control. They are classified as class II insecticides according to the WHO 2010 and are very toxic with a small margin of safety when they applied at low concentrations as individual compounds. However, the use of permethrin and piperonyl butoxide in a single formulation shampoo, such as in PAPA Plus and Licesol (Table 3), indicates the use of a chemical mixture. In this case, enhanced toxic effects may appear and expose children to possible poisoning. Nevertheless, several authors revealed the synergistic effects of piperonyl butoxide and permethrin and piperonyl butoxide and organophosphates (e.g., malathion) against insects. These compounds have been shown to cause health problems among children (Table 2). A high percentage of elevated levels of AST and urea concentration and reduced levels of AChE were also found among many children at different schools. From the toxicological point view, an elevated level of AST indicates oxidation stress on liver. Furthermore, elevated levels of urea in the blood serum indicate potential impairment of kidney cells. In contrast, reduced levels of AChE indicate potential cholinergic symptoms. These data indicated that health consequences were
present from the frequent application of medical treatments, in accords with previous reports\textsuperscript{35,38}. These data are also supported by the detection of malathion in urine samples of the infected students. Malathion is an organophosphorus insecticide. It is well known from the literature that organophosphorus insecticides are strong inhibitors of acetylcholinesterase and can cause immediate death at high levels of exposure. Additionally, Arslan et al.\textsuperscript{39} reported a genotoxic and cytotoxic potential for mixtures containing piperonyl butoxide. Our results agree with a previous report\textsuperscript{32} that revealed the decreased safety and efficacy of medical treatments for head lice. Meister and Ochsendorf\textsuperscript{40} observed the decreasing efficacy of permethrin and reported the successful treatment of head lice with ovicidal dimethicones. Medical treatment remains a problematic issue because the commonly used compounds are active against the mobile phase of insects (adult and nymph) without any effects of the immobile phase (eggs). Furthermore, these compounds have short residual effects (1-2 days); accordingly, the eggs that hatch after treatment can receive lower concentrations of active compounds and become more tolerant and eventually resistant to the medical treatment. Under this condition, the medical treatment becomes harmful to the children.

A comparison of our results with other studies shows the unique aspects of this study. This includes prevalence in different schools, density of insects per head, a liver biomarker, a kidney biomarker, a nervous system biomarker, toxicological symptoms, pesticide residue analysis in urine samples and surveyed medical treatments. Other studies\textsuperscript{9,13} examined prevalence of infestation, density of insect/head or medical treatment. In conclusion, the present study highlights the toxicity parameters of the medical treatments used in head lice control.

<table>
<thead>
<tr>
<th>Cluster zone</th>
<th>Average ± Standard deviation</th>
<th>Description of location</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Student/school</td>
<td>Infected cases/school</td>
</tr>
<tr>
<td>Rafah south</td>
<td>214± 35</td>
<td>22±5</td>
</tr>
<tr>
<td>Rafah north</td>
<td>204± 46</td>
<td>25±5</td>
</tr>
<tr>
<td>Khan Yonis east</td>
<td>223± 66</td>
<td>32±3</td>
</tr>
<tr>
<td>Khan Yonis west</td>
<td>255± 36</td>
<td>19±7</td>
</tr>
<tr>
<td>Middle zone</td>
<td>239± 45</td>
<td>43±3</td>
</tr>
<tr>
<td>Gaza</td>
<td>252± 46</td>
<td>18±4</td>
</tr>
<tr>
<td>Jabbalia</td>
<td>262± 61</td>
<td>27±7</td>
</tr>
<tr>
<td>Bait Lahia</td>
<td>277± 12</td>
<td>37±8</td>
</tr>
<tr>
<td>Bait Hanon</td>
<td>224±35</td>
<td>22±3</td>
</tr>
</tbody>
</table>
Table 2. Prevalence of infected cases, % infection, and average number of lice, nymphs and eggs/head among schools. Elevated case of AST, urea, reduced cases of AChE, positive samples of malathion residues in urine and frequency of shampoo application. Data in the same column that have similar numbers are not significantly different at a p-value = 0.05

<table>
<thead>
<tr>
<th>School name</th>
<th>Total students</th>
<th>IP</th>
<th>% of infection</th>
<th>Average ± standard deviation</th>
<th>Elevated case</th>
<th>Reduced case</th>
<th>Positive Malathion residue</th>
<th>Frequency of application</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>lice/head</td>
<td>nymph/head</td>
<td>eggs/head</td>
<td>AST</td>
<td>urea</td>
</tr>
<tr>
<td>Gaza A</td>
<td>357</td>
<td>32a</td>
<td>8.96</td>
<td>a8.7±0.58</td>
<td>a3.33±1.5</td>
<td>a5.3±2.5</td>
<td>7</td>
<td>3</td>
</tr>
<tr>
<td>Gaza B</td>
<td>467</td>
<td>27a</td>
<td>5.78</td>
<td>b5.3±1.5</td>
<td>a4.3±1.15</td>
<td>a6.3±2.3</td>
<td>5</td>
<td>2</td>
</tr>
<tr>
<td>Jabbalia A</td>
<td>793</td>
<td>10b</td>
<td>1.26</td>
<td>b4.7±1.5</td>
<td>a2.3±0.58</td>
<td>b2.3±0.6</td>
<td>6</td>
<td>1</td>
</tr>
<tr>
<td>Jabbalia B</td>
<td>670</td>
<td>30a</td>
<td>3.94</td>
<td>a9.3±0.6</td>
<td>a1.7±1.15</td>
<td>c3.7±2.1</td>
<td>4</td>
<td>0</td>
</tr>
<tr>
<td>Bait Lahia A</td>
<td>480</td>
<td>50c</td>
<td>10.41</td>
<td>a9.0±1.0</td>
<td>b6.3±1.53</td>
<td>d9.3±0.6</td>
<td>2</td>
<td>0</td>
</tr>
<tr>
<td>Bait Lahia B</td>
<td>586</td>
<td>15d</td>
<td>2.55</td>
<td>b5.7±0.6</td>
<td>a3.3±1.53</td>
<td>c3.7±1.2</td>
<td>3</td>
<td>2</td>
</tr>
<tr>
<td>Bait Lahia C</td>
<td>646</td>
<td>7b</td>
<td>1.08</td>
<td>c3.7±1.15</td>
<td>a2.3±0.6</td>
<td>b1.7±0.6</td>
<td>5</td>
<td>1</td>
</tr>
<tr>
<td>Um Anasser</td>
<td>466</td>
<td>20d</td>
<td>4.29</td>
<td>d6.67±2.1</td>
<td>b5.3±2.5</td>
<td>a6.7±1.5</td>
<td>2</td>
<td>1</td>
</tr>
<tr>
<td>Bait Hanon</td>
<td>624</td>
<td>22d</td>
<td>3.52</td>
<td>d7.0±2.7</td>
<td>a3.7±1.2</td>
<td>c3.3±0.6</td>
<td>1</td>
<td>2</td>
</tr>
</tbody>
</table>

Table 3. Traditional name, active ingredient and LD50 of chemical control compounds

<table>
<thead>
<tr>
<th>Traditional name</th>
<th>Active ingredient</th>
<th>LD50 mg/kg</th>
<th>Use</th>
</tr>
</thead>
<tbody>
<tr>
<td>TIC /TAC</td>
<td>Permethrin</td>
<td>430-4000</td>
<td>Three times/week</td>
</tr>
<tr>
<td>Lice Buster</td>
<td>Dimethicone 92%</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Cyclomethicone</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Chamomile oil</td>
<td></td>
<td></td>
</tr>
<tr>
<td>PAPA PLUS</td>
<td>Permethrin 1 g</td>
<td>430-4000</td>
<td>Two-three times/week</td>
</tr>
<tr>
<td></td>
<td>Malathion 0.5 g</td>
<td>1375-2800</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Piperonyl butoxide 4 g</td>
<td>7500</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Isododecane 94.5</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Licesol 100 mL container</td>
<td>Permethrin 0.155</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Piperonyl butoxide 1.65</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
CONCLUSION
A high prevalence of head lice (14.2% and 12.1%) was found in primary schools in middle zone and Khan Younis, respectively, whereas a low prevalence was observed in secondary schools (2.68%). The density of insects (adult, nymphs and eggs) on the students’ heads was found in the highly infected areas. The interesting outcomes of this study are the elevation of AST and urea levels and reduction in AChE in blood sera of the infected cases and malathion residues in urine samples that intensively used the medical treatments. It can be concluded that medical
treatment has health consequences among infected cases.

**SIGNIFICANT STATEMENT**

Head lice is an ectoparasite of public health importance around the world. The frequent use of medical treatments resulted in losing the biological activity of the active ingredient against head lice, creating tolerant and resistant species, and toxicological symptoms among children. We recommend researchers to test physical means, natural products and other safe and environmentally friendly products to control head lice infestation among children.

**Acknowledgments**

Prof Dr El-Nahhal would like to thank his students at Faculty of Science for helping with field and lab work. Special thanks to the Alexander von Humboldt Foundation for a Research Fellowship.

**Funding**

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**Ethics approval:**

This study was reviewed and approved by the Research Committee of the Faculty of Science and Medicine Research Ethical Committee. Formal approval was obtained from the Directorate of Health and Education, directors of the schools, class teachers, and the children’s parents. The study was conducted after explaining the aim of the study, and confidentiality was assured. Verbal consent was obtained from the children’s parents before examination. All children had the right to reject participation in the study.

**Contributor ship:** YE designed the study, actively participated in field work among schools, supervised blood sampling, and wrote and revised the manuscript. SH collected the photos, participated in analyzing the data, performed statistical analyses and obtained verbal consent from the children’s mothers.

**REFERENCES**


