

## CASE REPORT

# Persistent head lice following multiple treatments: Evidence for insecticide resistance in *Pediculus humanus capitis*

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### SUMMARY

Viable head lice were found on the scalps of two family members following multiple topical insecticide treatments. The possibility of reinfestation had been reliably excluded. Persistent infestation could be diagnosed only after cutting the hair and combing repeatedly, which allowed visualization of juvenile (nymphal) and adult lice. Insecticide-resistant headlouse infestations are probably much more common than is generally realised and may persist unnoticed, so that more aggressive approaches will be needed to eradicate these ectoparasites from individuals and communities.

**Key words:** lice infestations, pediculicide, pediculosis capitis, scalp dermatosis.

### INTRODUCTION

Pediculosis capitis, infestation of the scalp with the head louse *Pediculus humanus capitis*, occurs worldwide and is reported most commonly in school children.<sup>1</sup> In Australia, standard pediculicides are based on malathion or pyrethroids. Low-dose pyrethroids, herbal formulations and crotamiton are also available, with claims of therapeutic efficacy. While resistance of head lice to several pyrethroids (including natural pyrethrum and synthetic permethrin), malathion and related compounds has been confirmed overseas<sup>1–4</sup> and suspected in Australia,<sup>5</sup> treatment failure in Australia is still usually attributed to misdiagnosis, poor application of pediculicides or reinfestation. We describe active infestation of two family members where careful assessment revealed that head lice survived the application of various topical insecticides.

### CASE REPORT

The parents (both health professionals) of two young girls, aged 18 months and 5 years, noticed increasing scalp itching, first in the older daughter (who had small, crusted lesions on the neck and over the ears), then in all family members. Treatment was initiated when lice were seen crawling on the outer scalp hair simultaneously in the mother and older daughter. At first count over 300 eggs were seen close to the scalp of the older child, with more further out on her long and curly hair, and many were seen on the mother's moderately thick hair. The younger child with thinner hair had only a few eggs, identified quickly by their firm and eccentric attachment to the hair (their viability was not assessed). The finding of undetermined eggs is not sufficient to diagnose an active infestation. The father with very short hair had loose, desquamative epithelial plugs that encircled the hair as do old egg sheaths, but no evidence of infestation. Diagnosis was confirmed by light microscopy.

Initial treatment was commenced with the intention to follow State government health department guidelines, that is apply over-the-counter (non-prescription) pediculicide of choice after visually detecting live lice, and reapply after 1 week for any surviving eggs. The parents were faced with decisions about who to treat, the use of towels etc. before they could begin, because the necessary accompanying steps were confused by contradictory advice in public and peer-reviewed literature and manufacturer's directions, regarding inclusion of all family members (when only two individuals were evidently infested), egg removal, methods and frequency of application and necessity for treatment of fomites during treatment.<sup>6–8</sup> In order to prevent reinfestation, the parents: (i) isolated, inspected and treated all family members simultaneously in the home (no other residents) until no live lice were found on anyone; (ii) prevented close contact between family members (hair-to-hair and hair-to-body contact were carefully avoided during nappy changes and affectionate contact); (iii) attempted to remove all eggs from hair; and (iv) treated, by hot washing and hot drying, all potentially contaminated fomites (including bedding, towels, clothing) daily during treatment, in a fashion that was at least equal to the most stringent published standards.<sup>7</sup> Hats were not used and only freshly cleaned clothing and linen were used after each treatment and inspection.

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Additionally, upholstered furnishings were avoided, the daughters' interactions were closely supervised, fingernails were inspected and kept short, and regular combs (to remove tangles) and fine-toothed 'nit-combs' (for egg removal) were brushed clean and steam-sterilized between individual use. Use of hairbrushes and hair care products was discontinued. Treatment was applied, with more than the recommended volume if necessary, to coat thoroughly the entire scalp, to below the hairline, and all strands of hair completely. Hair was rinsed well after the required application time.

Because of the shortcomings of the standard topical treatment, despite the above precautions, the parents concluded that more treatment was necessary. The active ingredients used over a 10-day period were (in chronological order): malathion 1%, on two consecutive days; pyrethrins I and II 0.175% and piperonyl butoxide 1.75% w/w (a synergist); bioallethrin 0.66% and piperonyl butoxide 2.64%; permethrin 0.2% with citrus oils; crotamiton 10%; pyrethrins 0.165%, with piperonyl butoxide 2% and benzyl alcohol 8.07% w/v; pyrethrins 0.165% and piperonyl butoxide 4.0%; and a registered topical insect repellent preparation containing diethyltoluamide, di-*n*-propyl isocinchomeronate and *n*-octyl bicycloheptene dicarboximide (used twice, even though not registered specifically for head lice treatment).

Following the first treatment, active lice were found throughout the day on both the mother and 5-year-old daughter, prompting a repeat application, after which yet more live lice were found. Topical treatments were then rotated but, despite repeated thorough application of these pediculicides in various carriers (shampoo and lotion), interspersed with the use of the antiparasitic crotamiton and, finally, experimentation with the insect repellent, active lice were found again daily (at post-treatment intervals of 20 min to 24 h) over the 10-day period. While visual inspection did not reveal lice each day, combing with a variety (metal and plastic) of fine-toothed nit-combs did; because of this it was decided to apply yet another topical preparation each following day. No lice, alive or dead, were ever found on the father or 18-month-old daughter.

On day seven of chemical treatment, visual inspection and combing of the hair in sections with the fine-toothed combs for 20 min in each individual did not produce any live lice, suggesting successful eradication. However, a further period of continuous, vigorous, but painful fine-toothed combing produced not only eggs but also more live lice, including adult stages, from both mother and older daughter (the originally infested individuals). This prompted the parents to cut all family members' hair to facilitate the combing. Using sterilized scissors, cutting was done daily in graduated stages with the goal to retrieve any hypothetically remaining lice within a reasonable time, and while using a plastic fine-toothed nit-comb (Lady Jayne, Cork International, Australia) that did not scratch the scalp or pull hair out excessively and could be steam sterilized. Only after the hair was thinned and drastically shortened (to approximately 2 cm for thick or curly hair, and 10 cm for very thin hair) could the comb pass in one-second sweeps through the full hair thickness, along the scalp and out past the ends of the hair, without tugging. This enabled louse removal within an empirically reproducible 20 min of combing after each failed treatment and also made combing

more comfortable for the children. For the older girl with severely shortened hair, longer but well-thinned tresses of hair were left near the temples to maintain a feminine style which, along with a hidden shaved undercut around the ears and nape of the neck, still allowed efficient combing. Her compliance was important to allow further inspection and treatment. The father and younger girl remained louse free; the haircuts and inspections confirmed that they were not occult sources of infestation for the mother and older daughter.

Because of the measures taken to prevent transmission, it was concluded that the lice persisting after treatment were not newly acquired but resistant. Therefore, when more live lice (although fewer than before) were found on days 9 and 10 following applications of the last topical treatments, the chemical approach was abandoned and daily combing continued until finally no more live lice were found on day 11.

In total, 151 live lice (127 nymphal stages, 24 adults; 72 from mother, 79 from older daughter), ranging from 0.9 mm to 3.1 mm in length, were collected, mainly by fine-toothed combing (some dead lice were also found and some were rinsed away during treatment). Because not only nymphal but also adult lice were recovered over the 10 days of successive treatments, despite retrieval of as many as possible on each preceding day, it became obvious that hair cutting and repeated combing enhanced detection of the evasive mature lice (some were seen or felt escaping the section being combed) and the immature stages. All lice were beyond the newly hatched stage, and viable (tested by their ability to crawl along a loose hair); some had blood visible in the gut. Seven unhatched eggs positioned on hair more than 10 mm from the scalp were removed and kept at ambient temperature (ranging 15–34°C) and at 70–80% relative humidity. Two cream-coloured, soft-bodied nymphs hatched 6 days later; they were only 0.58 mm long, and crawled actively along hairs during observations over 12 hours.

Several lice that appeared dead or moribund (some of which were well-hidden in the comb slots or under the fingernails



**Figure 1** Six days following head lice bites to the skin showing inflammation and vesiculation.

after deliberate scalp-scratching) became active when brushed with a loose hair, climbing along it to the hand and then blood-feeding from the skin. Inflamed, exudative lesions developed, without scratching (Fig. 1). Biopsy by the family's general practitioner histologically confirmed a marked allergic reaction with deep lymphocytic and eosinophilic infiltration.

The mother and daughter experienced intense but diminishing scalp itching until no more live lice were found after the 10 days of chemical treatment. Subsequently, hair was kept short and fine-toothed combing was continued daily. Despite confirmed, persistent active pediculosis among close contacts when school resumed, no infestation occurred. The family notified all contacts; interviewing of contacts and others indicated that head lice first appeared in families after the children entered the early childhood or school community and that pediculicide rotation and experimentation with physical methods to detect and eliminate refractory infestations is widespread.

## DISCUSSION

The systematic removal, by combing, of a developing head-lice population is a recognized primary therapy.<sup>9,10</sup> When powerful, toxic and persistent insecticides such as DDT (dichloro-diphenyl-trichloroethane), used originally to control devastating louse-borne diseases, were extended to treating head lice they replaced the traditional approach of hair cutting and combing.<sup>11</sup> Lower concentrations of alternative pediculicides have since been introduced but their incomplete ovicidal activity may necessitate reapplication one week later to kill newly hatched nymphs. Effective treatment should kill at least all the walking stages of lice and ideally as many eggs as possible.<sup>12</sup> A typical suggested course of treatment involves two or even three applications of one pediculicide at weekly intervals, on the assumptions that only eggs might survive and that newly hatched lice are non-transmissible for 7 days.

In this study, active mature lice as well as nymphs were found on the two originally infested individuals after multiple treatments, but never on the other two family members. However, visual inspection and fine-toothed combing, although moderately sensitive, failed to detect the entire parasite population on either of the infested individuals. Only drastic hair-cutting followed by meticulous fine-toothed combing revealed remaining lice. This challenges the general belief that reinfestation or misapplication of pediculicides, accounts for lice found on schoolchildren after treatment<sup>15-15</sup> or after product trials.<sup>16,17</sup>

Head-lice infestations have increased worldwide despite the availability of pediculicides.<sup>5,18</sup> Failure to control head lice in communities has been attributed to erratic application of pediculicides<sup>8,14</sup> but, more recently, the effectiveness of these chemicals has been doubted. A systematic review and further analytical attempts have elicited limited evidence for clinical efficacy of topical treatments.<sup>19-21</sup>

More precise and reproducible methods of testing the effects of old and new treatments, clinically and *in vitro*, on both lice and their eggs, are required.<sup>18</sup> Moreover, the building evidence of resistance further underlines the need for regional testing to confirm suitability of treatments against local lice.<sup>18,20,22,25</sup>

The insect population is not only likely to develop resistance to broadly marketed over-the-counter products,<sup>2,20</sup> it may already, consequent to previous exposure to other insecticides, be insusceptible to another pediculicide tested elsewhere.<sup>18</sup> Indeed, where anecdotal reports of insecticidal failure in the field have been investigated in the laboratory, resistance of the lice and cross resistance to previously unused products have been confirmed.<sup>1,5</sup> High prevalence of head lice and consequent heavy usage of insecticidal products may explain the rapid selection of resistant strains,<sup>1,2</sup> with resistance mechanisms similar to those seen in other insect pests.<sup>1,5</sup> Sub-lethal doses of residual pediculicides may also encourage resistance;<sup>24</sup> so too may products that are underdosed or used for prophylaxis.<sup>19</sup> Increasing the dosage or using synergists does not necessarily overcome resistance,<sup>2,4</sup> nor is rotation of compounds agreed to be helpful.<sup>20,24,25</sup> Adequately controlled *in vitro* resistance testing is difficult<sup>5</sup> and clinical trials are costly and time-consuming.<sup>26</sup> In the absence of sensitivity testing in Australia, pediculicides have been rotated over the years in an attempt to minimize the development of resistance<sup>27</sup> although, as our findings indicate, this strategy has not been successful.

Patients with persistent infestation may have resistant lice or repeated infestation and need further assessment. Distinguishing these conditions may be difficult and depends on examination for active infestation after treatment but before potential re-exposure. Effective pediculicides should kill all lice within, or soon after, directed application times.<sup>26</sup> In one landmark study, a rapid killing effect of commercial products tested *in vitro* (on head lice with no prior exposure to pesticides) was reflected in clinical trials in which no live lice were found 15 min after application.<sup>26</sup> It can take more than 20 min of careful inspection to detect a few lice, which may be missed altogether in longer hair and possibly any thick or wavy hair that impedes straightforward combing; hidden by layers of hair, the lice themselves can escape disturbances.<sup>18</sup> Consequently, a treatment presumed to have been successful might have only reduced the louse population, so that inevitable relapse within 1 month or more later (the time taken for the remaining lice to grow through two generations) will be wrongly attributed to reinfestation. In this study, the risk of re-exposure was eliminated; live lice persisting on the same two individuals indicated multiple resistance.

The fact that a time-consuming, painstaking approach was needed to detect and remove these lice, contradicts authoritative advice, that regular grooming<sup>27,28</sup> or good hygiene<sup>29</sup> may discourage lice, even in longer hair. It was previously suggested that higher infestation rates were not actually related to longer hair, that cutting hair had no place in the control of pediculosis capitis and that, rather, brushing or combing could keep children relatively free of lice because it damaged lice.<sup>28,30</sup> Nevertheless, it was conceded that hair kept under approximately 8 mm would be less likely to support an infestation.<sup>28</sup> While the infirm, destitute or indifferent may neglect to treat an established infestation, daily modern grooming (e.g. washing and brushing of hair as carried out by the family in this study before diagnosis) has not been shown to prevent or cure head lice. The historical success of shaving, the lice' behaviour of not always remaining confined to the scalp but

wandering through the hair, and the easier examination of short, fine hair with the possibility of skewed epidemiological results,<sup>2</sup> together with our observations, support an older expert assertion that, the heavier the hair, the higher the infestation rates and the more difficult or unreliable the task of physically detecting or eradicating lice.<sup>11,15</sup>

Misdiagnosis of dead or empty eggs on outgrowing hair is also blamed for unnecessary treatment or imaginary persistent lice.<sup>4,51,52</sup> Egg removal has been discouraged<sup>8</sup> but this confuses the clinical picture, as published criteria for identification of viable eggs are inconsistent.<sup>14,53</sup> New eggs laid close to the scalp-hair junction after treatment indicate ongoing activity, but excluding an active infestation requires more care. Obviously, flat empty egg-casings are non-viable, but visual, even microscopic, identification of viable eggs in a heavy infestation, on the basis of appearance<sup>54</sup> or arbitrary distances of up to 6–10 mm from the scalp,<sup>25,32</sup> may be impractical. In the present case report, unhatched eggs found further along the hair shafts from the scalp were shown to be viable by hatching experiment. Outgrown eggs may be markers of either past cleared or possibly ongoing infestation; closer and repeated follow-up inspections may be needed to avoid false-negative findings.

The finding of scant eggs on the younger girl at the outset of our study needs explanation. Perhaps she had only recently acquired a small number of adult lice, too few to be observed visually but enough to lay detectable eggs. Alternatively, her thin hair may not have supported the establishment of invading lice, despite her close contact with actively infested family members prior to their diagnosis and treatment.

The experience of this family illustrates the inadequacy of clinical assessment of products by visual inspection<sup>18</sup> and the need for a more reliable screening technique. Optical equipment<sup>14</sup> and fine-toothed combing to detect resistant lice<sup>55</sup> have been recommended, but this study suggests that, while they may be preferable to visual inspection alone, they may also have limitations during one-off inspections. The challenge to find every louse, from newly hatched nymph to adult, among over 100 000 hairs is daunting! Furthermore, any louse found must be checked carefully for viability if assessing chemical treatment.

Formal, regular screening, recommended for the control of head lice,<sup>2,56</sup> is not routine practice in most Australian schools; responsibility appears to rest with individual families and, occasionally, their doctors. The National Health and Medical Research Council (NHMRC) recommends that children with live lice be excluded from school until standard treatment is commenced but, in the absence of classroom screening, children with resistant lice, repeat or light infestations will be overlooked.

Reliable evidence supporting either chemical or alternative treatments is lacking and advice is conflicting.<sup>21,34</sup> Recommendation to use pediculicides repeatedly cannot be supported because, despite current public health advice to use head lice treatment products if live lice are found, uncut hair may hide resistant lice, and such insecticidal products should not be used for long periods. Consumers, misled by some manufacturers' claims of complete efficacy, may assume recurring lice to be a failure on their part, or a reinfestation

from others and so lock into a cycle of ineffective pediculicide treatment. We do not propose that the laborious measures to prevent reinfestation used by the family of this report are necessary; rather, their controlled regimen allowed us to conclude that incompletely eradicated *P. humanus capitis* could be elusive and difficult to cure completely, as has been suspected in the wider community.

Our report of a measurable hair threshold (dependent on individual hair type and scalp sensitivity rather than nominal hair length) that facilitates efficient combing for systematic louse removal, could be the starting point for evaluation of hair factors in this area. Combing of various methods provides a non-toxic approach that should be encouraged.<sup>9–11, 54</sup> Fine-toothed combing to detect and remove head lice is made more efficient by shortening and thinning the hair, which also minimizes discomfort and improves cooperation from young children.

Louse infestation can be distressing and costly to families, and can have complications; dangerous or flammable chemicals have seriously injured children,<sup>54</sup> while chronic irritation and secondary infection<sup>11,37,58</sup> may disrupt behaviour and school performance.<sup>30,59</sup> The vectorial potential of head lice remains unresolved; they might transmit typhus<sup>40–42</sup> and louse-borne diseases are re-emerging in both developing and developed countries.<sup>45</sup> Although head lice have been referred to as a social nuisance,<sup>15,25</sup> their resurgence warrants closer surveillance and research.

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