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A Novel Nit Comb Concept Using Ultrasound Actuation: Preclinical Evaluation

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Abstract

Nit combing and removal of head louse, *Pediculus humanus capitis* De Geer (Anoplura: Pediculidae), eggs is a task made more difficult because "nit combs" vary in efficiency. There is currently no evidence that the binding of the eggshell to the hair can be loosened chemically and few hair treatments improve the slip of the louse eggs along the hair. Ultrasound, applied through the teeth of a nit comb, may facilitate the flow of fluids into the gap between the hair shaft and the tube of fixative holding louse eggs in place to improve lubrication. Ultrasound alone had little effect to initiate sliding, requiring a force of 121.5 ± 23.8 millinewtons (mN) compared with 125.8 ± 18.0 mN without ultrasound, but once the egg started to move it made the process easier. In the presence of a conditioner-like creamy lotion, ultrasound reduced the Peak force required to start movement to 24.3 ± 8.8 mN from 50.4 ± 13.0 mN without ultrasound. In contrast, some head louse treatments made removal of eggs more difficult, requiring approximately twice the Peak force to initiate movement compared with dry hair in the absence of ultrasound. However, following application of ultrasound, the forces required to initiate movement increased for an essential oil product, remained the same for isopropyl myristate and cyclomethicone, and halved for 4% dimeticone lotion. Fixing the nit comb at an estimated angle of 16.5° to the direction of pull gave an optimum effect to improve the removal process when a suitable lubricant was used.

Key words: head lice, *Pediculus humanus humanus*, treatment, nit removal, ultrasound

Removing head louse, *Pediculus humanus capitis* De Geer (Anoplura: Pediculidae), eggs and the empty eggshells (nits), which remain attached to hairs long after the nymphs have emerged, is a difficult process. For most people, the presence of nits is mainly a cosmetic concern, being unsightly and potentially stigmatizing evidence of recent infestation. However, in some jurisdictions, louse egg and nit removal can be an important policy issue resulting in exclusion of children from school, with associated costs to schools and households in terms of disruption, stigmatization, and loss of income (Greene 1898, Mumcuoglu et al. 2006).

Historically egg and nit removal has mainly been through some form of combing (Mumcuoglu 2008). But, so-called "nit combs" are so extremely variable in both construction and design that many are not fit for purpose, and remove neither eggs nor nits because the teeth are too widely spaced (Larsen and Burgess 2002, Mumcuoglu 2008, Gallardo et al. 2013). In contrast are combs that have teeth that are set into the base in such a manner that they damage the hair by shredding it or pulling it out because the gap between the teeth is too narrow (Larsen and Burgess 2002). As a result most people find nit combing highly onerous, a task that is made more difficult because most viable eggs are not easy to detect, being fixed to the hair close to the scalp.

Attempts to make nit removal easier by chemically altering the bond between the cylinder of louse egg fixative and the hair are unsuccessful because there is no detectable chemical interaction between the fixative and the hair shaft (Burkhart et al. 1999) and we have previously shown that several products claiming to facilitate removal were no more effective than normal hair conditioner or even water alone (Burgess 2010). The same study also found that when a louse egg is pulled along a hair the tube of fixative tends to rotate around the hair shaft. On round hairs this made no difference to the force required but on oval hairs this resulted in an increase in tension as the narrow cross section of the fixative tube twisted over the wider diameter of the hair. Consequently, we concluded that any mechanism that could reduce this "locking" of the fixative would be beneficial when removing nits.

Since it was difficult to identify a fluid that made sliding nits easier, it was postulated that if a fluid could be induced to flow into the space between the fixative and the hair surface it should be easier to remove the eggs. Making a viscous and thixotropic fluid like hair conditioner flow more readily is not easy. However, the rheological characteristics of the material could be altered by application of ultrasound at the right frequency to cause cavitation effects.

This would allow the emulsion to develop better flow characteristics over short distances.

The investigation was conducted as part of a co-operative research project within the Sixth Framework Programme of the European Commission. It aimed to develop a louse egg removal system with a purpose-designed comb using ultrasound to alter the flow characteristics of various fluids, which could make louse egg and nit removal easier.

Materials and Methods

Louse Egg Removal

We obtained louse eggs on hairs by supplying recently fed laboratory-reared human body/clothing lice, *Pediculus humanus humanus*, with locks of washed, unbleached, human hair over a 48-h period. The eggs of clothing lice are of a similar size to those of head lice but the female louse generally fixes these to the substrate using a smaller volume of the adhesive fluid than a head louse. This probably means that clothing louse eggs can be removed from hairs more easily than head louse eggs, but the advantage of using eggs from laboratory clothing lice was that a large number of eggs of the same age and general quality could be obtained relatively quickly in a repeatable manner. These were then used to measure the force required to remove louse eggs from hairs using a modified version of the technique described previously, in which individual hairs bearing louse eggs were pulled by a slip-peel tester (SP-2000, IMASS, Inc., Accord, MA) through a fixed glass microtube to displace the eggshells (Burgess 2010). In this series of tests the glass microtube, described previously for holding the louse eggs, was replaced by a metal comb with teeth close enough that they could remove louse eggs and nits from hairs. Combs used in the tests were fixed to the end of the platen of the slip-peel tester using PVC tape so that the teeth were held vertically. The alignment of the comb could be adjusted relative to the direction of movement by placing wedges between one end of the comb and the vertical face of the platen. For each test series a minimum of 20 louse eggs were employed.

The first stage of the investigation required baseline data for each of the measurements of force required to initiate and continue sliding of the louse eggs along hairs using untreated hairs and eggs in a dry state. These were the "Peak force," the force required to break the physical grip of the fixative tube holding an individual louse egg in place and to initiate sliding, and the "Average force." In this context the term "Average," as given by the digital readout from the device, is not a mean or averaging figure calculated from the results of several separate tests but is the average force encountered by a single eggshell as it slides, or judders as the case may be, along the hair shaft, including any effects incurred by twisting of the hair shaft within the fixative tube (Burgess 2010). The same force measurements were also made after subjecting separate batches of untreated eggs to ultrasound.

Ultrasound Delivery

In order to produce a lightweight device that was efficient at transmitting ultrasound efficiently, an aluminum lever was coupled with the nickel silver comb unit chosen for inclusion in the final device (Innomed comb, Hogil Pharmaceutical Corp, White Plains, NY) to form a unit with a lateral dimension equivalent to the wave length of the ultrasound. This allowed optimum energy use from the least power source.

For the initial bench study, ultrasound was delivered using an ultrasonic processor for small volumes (VCX134PB, Sonics & Materials, Inc., Newtown, CT), which was capable of delivering 134 watts at 20 kHz. The initial experiments on the effects of ultrasound on head louse treatment fluids were conducted using the supplied stepped probe but later experiments for measurements of the direct effects of ultrasound on fluid penetration into the louse egg fixative tube used a comb with a heavy stainless steel base and robust pins that screwed onto the probe attachment supplied with the processor.

Data from the development process and comb experiments were used in preparation of the ultrasound converter for a prototype nit comb device powered by four AA/R6 size 1.5 volt batteries. The prototype, which was constructed using a 3-D printer, operated using two piezoelectric actuators delivering an axial stroke of the comb of ~0.23 μm at 35 kHz (equivalent to 0.051 m/s of ultrasound generation) and a transverse stroke measured between 0.12 and 0.66 μm at 35 kHz (equivalent to 0.02 to 0.145 m/s of ultrasound generation). These data were obtained using a dedicated test rig by the specialist subcontractor to the project (Cedrat Technologies S.A.) and summarized in a subsequent report (Innopol 2007). The latter value depended upon where the comb unit was located along the housing. The back plate of the leverage unit actuated 0.2 μm at 35 kHz, which suggested that the thermoplastic elastomer seal holding the actuator unit in place was constantly under compression when the leverage subsystem was powered up. This, together with some stiffness in the leverage and housing, provided a certain amount of damping effect that needed to be minimized in the clinical version to improve efficiency (Innopol 2007). The prototype incorporated a switch system with linked LEDs, one to indicate that the battery power remained within specification and the other to show that the ultrasound actuators were functioning.

Combs

Two types of comb were used. The first was a heavy stainless steel comb, which was an excellent conductor and provided an optimum delivery of ultrasound to the louse eggshells and any fluids applied to the hairs. The data obtained from that comb were used to check compatibility of the later development work using the nickel silver comb unit, which was selected because the teeth were of high quality metal with a consistent profile, welded into a phosphor-bronze strip, giving considerable rigidity and a consistent tooth gap across each individual comb unit, although units manufactured at different times show some variation in the average tooth gaps. Measurements of the gaps between 20 consecutive teeth from three combs each manufactured about 10 yr apart, from the mid-1990s onward, were 210 μm (*SEM* 4.22 μm), 238 μm (*SEM* 4.73 μm), and 165 μm (*SEM* 2.95 μm).

Lubricant Fluids

We investigated several preparations for their ability to facilitate louse egg removal from hair with and without exposure to ultrasound. The product that was expected to produce the best results was a conditioner base containing 1% neem oil (Nice 'n Clear head lice lotion, Nelsons, Wimbledon, United Kingdom). Based on previous work with conditioning rinses, this product was considered to be potentially suitable for identifying any increased penetration of fluid into the gap between the hair and the nit fixative under the influence of ultrasound induced cavitations. Comparison products were selected from other widely available physically acting pediculicides including 4% dimeticone lotion (Hedrin 4% lotion, Thornton & Ross Ltd, Huddersfield, United Kingdom), isopropyl myristate and cyclomethicone (Full Marks solution, Reckitt Benckiser Plc, Slough, United Kingdom), and an essential oil in vegetable oil mixture that claims to facilitate louse egg removal (Nitty Gritty aromatherapy head lice solution, Oakwood Remedies, London, United Kingdom). The effect of each of these products to reduce the resistance to sliding by the louse egg was compared with the forces required to move untreated eggs on dry hairs.

The method employed groups of hairs ∼15 cm long bearing louse eggs that were separated from a larger hair tress and allocated to marked Petri dishes ready for subsequent exposure. Each hair tuft contained up to 50 hairs. Immediately prior to testing, each tuft of hair was immersed in the appropriate fluid for 10 min. After exposure the excess fluid was blotted off and individual hairs selected on which the egg was situated closer to one end, with the operculum pointing toward the shorter end. The forces required to remove at least 20 eggs were then measured.

Analyses

Force data obtained using each of the test procedures were compared with the appropriate control batch by means of nonparametric analyses, using the Kruskal–Wallis and Mann–Whitney tests. We conducted analyses using Oxstat II (version 1.1; Oxstat Ltd, London, United Kingdom) and a purpose built calculator for Kruskal–Wallis/Mann–Whitney tests (University of Delaware 2008).

Results

The initial stage of testing using eggs on dry and untreated hairs found that ultrasound had only a marginal effect on both the Peak and Average force measurements, indicating that ultrasound alone did not detectably facilitate sliding of the egg along the hair shaft (Table 1).

Of these treatments, the 1% neem conditioner formulation was the only one found to reduce the mean Peak force below that obtained using dry hair ($P = 0.007$). Each of the others appeared to cause a significant ($P < 0.02$) increase in Peak force (Table 1). However, in all cases there was little to choose between the levels of Average force, although the conditioning base appeared to reduce friction to some extent. When ultrasound was applied to each of the treated batches of hair for 10 s there was a reduction of mean Peak force in those eggs treated using Nice 'n Clear ($P = 0.014$) and Hedrin 4% lotion ($P = 0.019$) but no change in those exposed to Full Marks solution ($P = 0.655$). Interestingly there was a significant increase ($P < 0.0016$) in the Peak force required to move eggs treated with the essential oil-based Nitty Gritty solution compared with the control batch of eggs (Table 1). Once movement had started, sliding of the eggs along the hairs was not significantly affected by the treatment.

Our previous comparison of nit combs had shown that it was possible for louse eggs and nits to slip between the teeth when single hairs were drawn straight through the comb (Larsen and Burgess 2002). By changing the angle of the face of the comb relative to the direction of pull, using wedges fitted between the back of the comb and the platen block and calculating the angle trigonometrically, found that an estimated angle of 16.5° was more effective to ensure all louse eggs were removed without causing the hair to either break or snag in the teeth of the comb than lesser angles, or greater angles up to 20°. On the dry hair sample the change of angle plus ultrasound was observed to increase the mean Peak force compared with the straight pull, although this difference was not significant ($P = 0.33$). However, when the eggs and hair were treated with the neem conditioner a relative reduction in both Peak and Average force values was found using the angled comb both without and with ultrasound when compared with the straight pull equivalents (Fig. 1) with a nonsignificant ($P = 0.099$) trend for reduced Peak force using the angled comb together with ultrasound.

Discussion

In this investigation, we have confirmed our previous findings (Burgess 2010) that a thixotropic emulsion similar to a hair conditioning rinse can help facilitate removal of louse eggs and nits from hair. We have also investigated some alternative widely used oily head louse treatments for the effect they have on nit removal. The results indicate that the low surface tension of siloxane chemicals such as dimeticone and cyclomethicone, which are among the most common ingredients in modern treatment preparations, although potentially increasing penetration of the fluid into the narrow space between the fixative and the hair, do not lubricate the hair surface to facilitate sliding of the egg along the hair. In contrast, the relatively viscous and high surface tension of a conditioning base, which was believed to exhibit cavitations under the influence of ultrasound, produced a good lubricant surface that appears capable of reducing the Peak force required to initiate sliding to the same level as is required for continued sliding of the eggs along the hair shaft (Table 1). However, in this series of comparisons, what was most striking was the increased binding of the egg fixative to the hairs when exposed to the essential oil mixture Nitty Gritty aromatherapy head lice solution, which, contrary to the marketing claims for the product, caused Peak force to increase by ∼75% under the influence of ultrasound. The essential oil components of the product appear to cause contraction of the fixative tube so that it grips the hair shaft more tightly (E. R. B., unpublished data).

The success of conventional nit combing appears to be limited by the quality of the devices used, and none appear to be as good as John Sacker's patented nit comb, which was made from solid brass with parallel sided teeth one hair width apart (Sacker 1942). Most modern nit combs are made using rounded metal pins, which may have teeth set close together but this placement is often irregular

Table 1. Peak and Average force measures required to remove louse eggs and nits by the prototype ClearBrush without and with ultrasound activation using different products as lubricants

Ultrasound	Lubricant	None ($n = 40$)	N 'n C ($n = 20$)	NG ($n = 20$)	FMS ($n = 20$)	H ($n = 20$)
Off	Peak force (*SEM*)	125.8 (17.95)	50.4 (13.04)	194.8 (60.31)	224.6 (24.91)	249.6 (35.99)
	Average force (*SEM*)	17.3 (2.35)	5.2 (1.18)	33.9 (11.08)	25.3 (8.43)	16.6 (3.82)
On	Peak force (*SEM*)	121.5 (23.83)	24.3 (8.83)	352.1 (91.69)	229.5 (30.89)	124.0 (39.72)
	Average force (*SEM*)	12.9 (4.02)	25.5 (0.49)	45.1 (16.57)	34.3 (6.77)	18.1 (12.55)

Force figures are given in millinewtons. Lubricants used were N 'n C—Nice 'n Clear head lice lotion; NG—Nitty Gritty aromatherapy head lice solution; FMS—Full Marks solution; H—Hedrin 4% lotion.

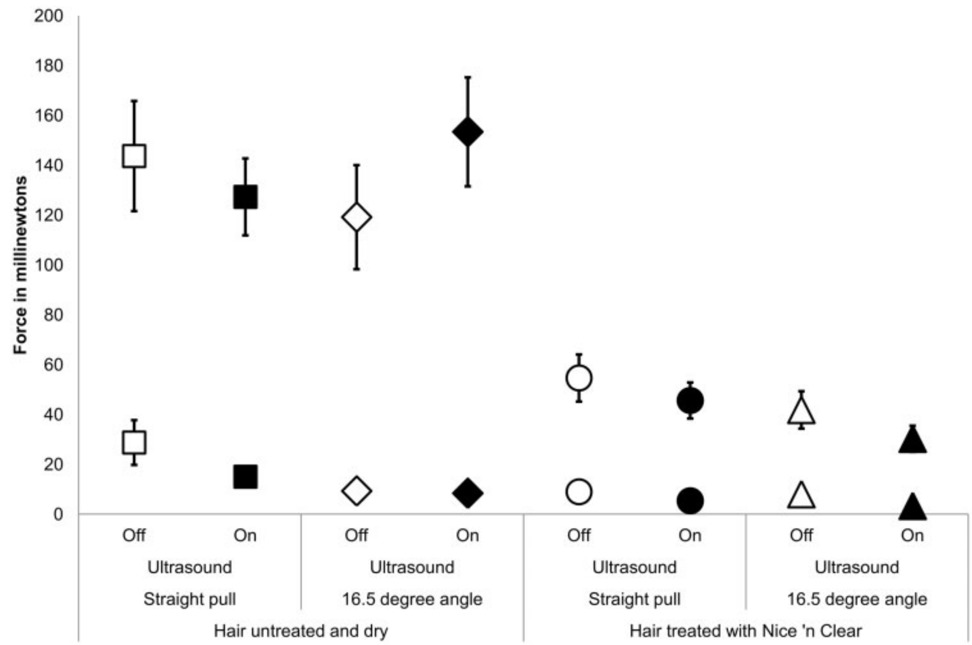

Fig. 1. Comparison of the Peak and Average force required to remove 20 louse eggs and nits using either a straight pull, or with the comb unit at an estimated angle of 16.5°, either with or without neem conditioner as a lubricant, using the ClearBrush prototype comb. Open symbols relate to use with ultrasound switched off, closed symbols to use with ultrasound applied. In each case the higher recorded figure (upper symbol) relates to the Peak force and the lower recorded figure (lower symbol) relates to the Average force. Error bars represent one standard error from the mean (*SEM*). Where error bars are not visible the *SEM* was sufficiently low the bars fell within the boundary of the symbol.

(Larsen and Burgess 2002) and even when set close together the empty eggshells are often compressed so that they are rolled between the pins without being removed (Burgess et al. 2010). The comb tooth unit used in this study avoids some of these problems by having teeth spaced closer than many other products and also having blunt faces to the teeth but even these characteristics did not prevent some eggs and nits from slipping between the teeth when the comb unit is held, in what could be considered a normal position, at right angles to the direction of pull on the hair (Larsen and Burgess 2002). We found that rotating the comb, so that the teeth were presented at a slight angle to the direction of pull, reduced the number of nits slipping between the teeth and artificially produced the same level of torque on the nits as would be produced by the teeth of Sacker's comb. Trial and error testing of different angles found that the efficiency of the comb to remove all nits increased as the angle increased up to the estimated 16.5° angle but declined above this because too much tension was applied to the hair as the angle increased. We found that by introducing a slight angle in the presentation of the comb the eggs or nits were held at the leading edge of the teeth of the comb and pulled along the hair shaft with minimal additional force, which in turn was minimized by addition of the conditioner-based lotion, especially when ultrasound was applied (Fig. 1).

The use of ultrasound for activation of fluids applied to hair is not a completely new idea, although previously it has been used for styling or application of fixative chemicals (Quan et al. 2003, Olshavsky and Quan 2004, Nunomura 2007). However, use of this technology for application and dispersal of head louse treatment products is a novel approach. The findings of this study having demonstrated the potential viability of the original hypothesis upon which the project was initiated, the next stage was to develop usable prototype devices for use in a clinical investigation, to be described in a separate report.

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